

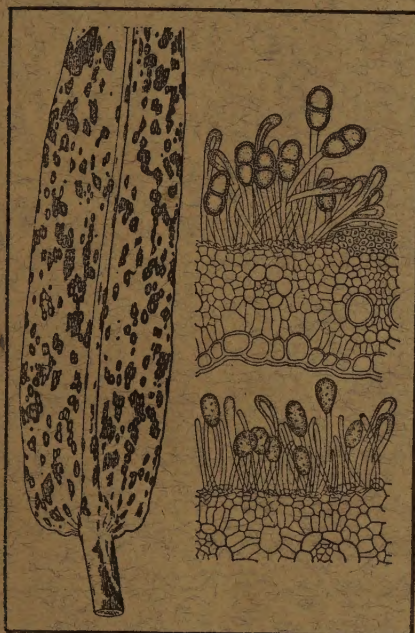
# INDIAN PHYTOPATHOLOGY

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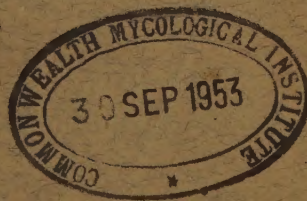
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Dedicated to

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1896—1952

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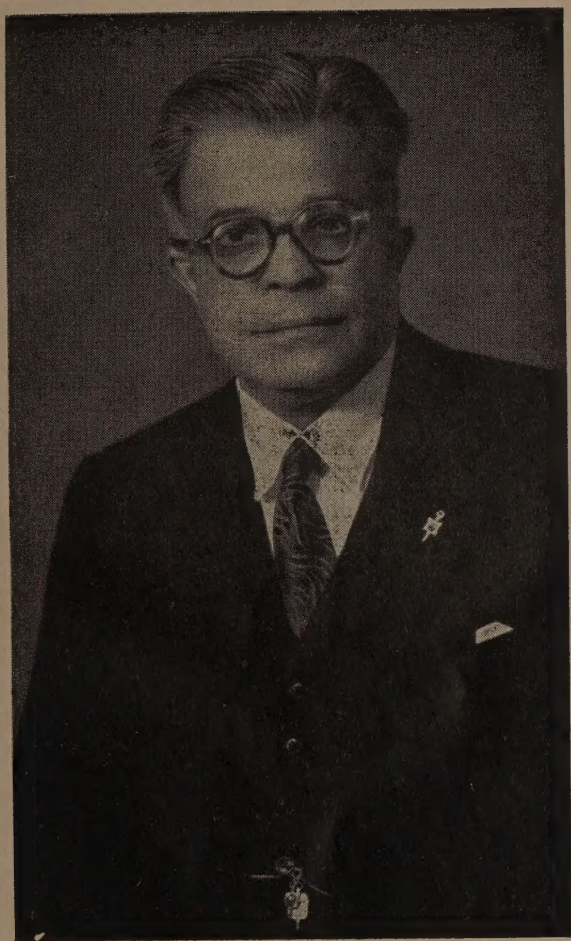
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## B. B. MUNDKUR

( 1896—1952 )

Balchandra Bhavanishankar Mundkur, Professor of Botany, Poona University, India, died on December 13, 1952, at the the age of fifty-six. He was born on June 26, 1896, in Mundkur, a village near Mangalore of the Madras State. After passing the high school examination in 1915, he had his early collegiate education at the St. Aloysius College, Mangalore. Later he studied in the Presidency College, Madras, from where he took B.A. (Hons.) in Botany with a first class and won a 'Gold Medal'. After his brilliant academic career, he was for a short time an Agricultural Officer in Bengal, but from 1922 onward his professional career was solely devoted to Mycology and Plant Pathology. From 1922 to 1928 he worked as Assistant Mycologist in the Cotton Research Scheme at Dharwar (Bombay State) and made a valuable scientific contribution on 'wilt disease' of cotton. This work laid a firm foundation for his future career.

Dr. Mundkur went to U.S.A. in 1929 for post-graduate studies at the Iowa State College of Agriculture and was awarded the Ph. D. degree in 1931 in Plant Pathology. Soon after his return he was appointed as Asstt. Mycologist in the Division of Mycology, Indian Agricultural Research Institute, Pusa, Bihar. He was associated with the Institution, both in Bihar and later at Delhi till 1947. During his tenure of the post, Dr. Mundkur was a popular figure with post-graduate students and members of the staff. In the earlier part of his career, he took a very keen interest in the 'Ustilaginales' of India. He also prepared a supplement to 'Fungi of India' by Butler and Bisby. The numerous post-graduate students, who worked under him, found him a wise guide and friendly councillor. He demanded from them the highest standard of scholarship, technical ability and assiduity. The capacity which Dr. Mundkur possessed to enthuse his students remains unrivalled. His devotion to his subject was exemplary. Dr. Mundkur had wide international contacts and possessed a valuable collection of reprints which he generously handed over to Fergusson college Poona. His professional advice was sought for avidly by mycological workers all over India and he never shirked in rendering every possible help to those who needed his help and guidance. His persistent effort was to raise the status of Mycology and Plant Pathology in India to that of a major subject in the Indian Universities and in the Agricultural Institutions. His publication of text book in 1949 entitled 'Fungi and Plant Diseases' was an endeavour in this direction.

In recognition of the meritorious service, in 1947 he was selected as the first Deputy Director of Plant Diseases in the newly established Directorate of Plant Protection, Quarantine and Storage of the Ministry of Food and Agriculture. He actively helped in the development of Plant Protection Services in the various States of India and in the development of Plant Quarantine work. His pioneering work in this field will be remembered with great gratitude, for this was the first serious effort to narrow the bridge between research and development work in India. He was becoming increasingly conscious that application of research was



essential as an aid in solving the food problem of India—a act which was projected with full force as a result of partition of India on the eve of her independence. Dr. Mundkur's health was failing but with single-minded devotion to his work, he extensively toured India, helping and advising in the development of plant protection work. This involved considerable amount of work which Dr. Mundkur undertook readily at great personal discomfort and in absolute disregard of his failing health. He was suffering from diabetes and high blood pressure and the strain on his health was too great.

After his retirement Dr. Mundkur was appointed Professor of Botany and was the first incumbent of this newly created post at Poona University. With his characteristic thoroughness he gave his undivided attention to his new assignment and organised excellent laboratory and teaching facilities. His health was rapidly falling and his numerous well-wishers and friends advised him complete retirement and rest. However, in his undaunted zeal to accomplish the work he had undertaken, no amount of advice or persuasion could sway him. He had a heart attack and was confined to nursing home for sometime at Poona. Soon thereafter he visited Delhi and met his numerous friends. His physical state showed ominous signs, but still his enthusiasm was unbounded. While at Delhi he discussed with the writer the various things which, he thought, needed the attention of Plant Pathologists in India. Future development of Plant Pathology and Mycology in India was foremost in his mind. His end came suddenly at Poona on 13th December 1952 and India lost a Mycologist and Scientist of outstanding ability.

Dr. Mundkur published about 80 papers covering a wide field. His last work 'Genera of Rusts' in joint authorship with Dr. Thirumalachar has been well received by Mycological workers and is soon to be published in the form of a book. He was one of the chief sponsors of the Indian Phytopathological Society and its organ 'Indian Phytopathology'. But for his inspiring lead the Society could not have come into existence in such a short time. He was twice President of the Society, the Editor-in-chief of its journal and Secretary-Treasurer. He was also connected with many other scientific societies in India and abroad.

Dr. Mundkur was a Fellow of the National Institute of Sciences of India, member of the American Phytopathological society and the society of the Sigma XI, and in 1951, presided over the Botany Section of the Indian Science Congress Session held at Bangalore.

He acted as a member of Agricultural Mission sent by the Government of India to Afghanistan in 1939. In 1947, he was the official delegate of Government of India to the Commonwealth Mycological Conference in London. In 1949 he represented the Government of India on the Rubber Conference held at Singapore.

The name of Dr. Mundkur will always be honoured as that of a man who pursued his life-work with acknowledged success, ceaseless devotion and singleness of purpose, and his death is lamented by his numerous friends and colleagues.

He leaves behind his wife and two sons, and to them we offer our most sincere sympathy in their loss.

Directorate of Plant Protection,  
Quarantine & Storage,  
New Delhi.

P. R. MEHTA.



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## SUBMUTANTS FROM NONSYSTEMIC MUTANT STRAINS OF TOBACCO-MOSAIC VIRUS

FRANCIS O. HOLMES

(Accepted for publication January 15, 1952)

In an earlier paper it was reported that a strain of tobacco-mosaic virus (*Marmor tabaci* H.) may mutate in such a way that additional mutants from this strain and submutants\* from a derivative constitute two series of variants differing in many respects among themselves but distinguishable, as series, by a character appearing in the first mutation and retained during subsequent changes (Holmes, 1936). Independent mutations in viruses have been reported also by Norval (1938), Hershey (1946), and Friedrich-Freksa *et al.* (1946). Kunkel (1934) previously had observed types of mutation that occurred sometimes simultaneously but sometimes separately; these were later recognized (Kunkel, 1947) as showing independence. Melchers (1942) reported parallel mutations within distinctive lines of tobacco-mosaic virus. Gowen (1945) noted that susceptibility to x-ray inactivation was not changed in mutants and suggested that this may mean that the size of the viral particle was not changed while mutation was taking place. Collectively these observations serve to indicate that viruses often change in one part of their genetic structure without obliterating characteristics acquired previously in other parts.

Independence of hereditary changes with respect to each other is not an invariable rule for tobacco-mosaic virus, however. In the present paper it will be shown that certain changes are so related as not to be independent of each other at all so far as has been observed.

In the course of the study earlier mentioned (Holmes, 1936), it was noted that both of the source strains, respectively known as the *distorting* and the *masked* strains of tobacco-mosaic virus, besides giving yellow-mottling variants that were distinguishable as groups by differences in ability to penetrate very young tissues at the top of the host plant, gave a considerable number of yellow-spotting variants that characteristically failed to move from the inoculated leaf. This class of nonsystemic variants was first reported in the literature by Jensen (1933). Such derivatives, because they rarely left the inoculated leaf, could not be compared with each other in respect to their potential tendencies to penetrate young tissues at the top of the plant. They differed among themselves in many characteristics, some producing yellow-spot lesions, others greenish-yellow-spot lesions, still others concentric ring markings, and so forth.

In general it has been the experience of those who have observed non-systemic strains of tobacco-mosaic virus, that these are of low apparent infectivity (Holmes, 1936; Jensen, 1933, 1937; Norval, 1938). Indeed, Norval

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\* The words *submutant*, *mutant*, *variant*, and *derivative* are used as equivalent in meaning in this paper, except that variants from the original strain of tobacco-mosaic virus are referred to as *mutants*, whereas variants derived from these mutants are referred to as *submutants* to indicate their origin.



(1938) compared the characteristics of three strains that differed in ability to move from the site of inoculation and noted a correlation between freedom of movement and infectivity.

It is the purpose of the present paper to record attempts at isolation of systemically spreading variants from two localized strains of tobacco-mosaic virus, one of which was observed to be much more unstable than the other and, hence, a better source of variants in the sense that more variants could be isolated for each unit of time and for each lot of infected plants. In the course of the experiments recorded here, it was found that a strict correlation existed between changes that produced variants capable of moving freely from the site of inoculation and changes that increased infectivity.

#### MATERIALS AND METHODS

A single-lesion isolate of the green-mottling "distorting" strain (Holmes, 1936) of tobacco-mosaic virus was used as a source strain to inoculate young Turkish tobacco plants (*Nicotiana tabacum* L.). From Atypical spots in the patterns of mosaic on leaves of these plants, usually yellow spots or partly necrotic spots, transfers of virus were made by pin-puncture to healthy plants of the same kind. Some of the plants thus inoculated showed yellow-mottling systemic infection and others merely localized infection.

Two mutant strains of virus that tended to remain localized were selected. One, number 1933B, was of an unusual type, producing lesions often ringed with delicate white necrotic lines and becoming yellowed only in late stages of infection; the other, number 1940F, was of a much commoner type, producing yellow-spot lesions only. Stocks of these two original nonsystemic mutant strains of virus were maintained in young plants of Turkish tobacco. They were transferred to new plants every ten to twelve days, always from plants not showing evidence of the presence of additional, *i.e.*, submutant, strains.

Manifestations of disease produced by new submutants were observed as they developed in inoculated plants of Turkish tobacco grown in 4-inch clay pots. The plants were propagated in a greenhouse that was maintained at about 22° C. at night, with day temperatures usually somewhat higher than this. Similar plants not inoculated, but growing near inoculated plants, consistently remained free from signs of disease.

Samples of submutant strains derived from the two original mutant nonsystemic stocks of virus were preserved in fragments of infected leaves. The leaf fragments were dried in cellophane tubes which were later stored in metal-clasp envelopes.

Measurements of infectivity of submutant strains and isolates were made by inoculating 5 leaves of a plant of *Nicotiana glutinosa* L. with each. Plants of this species were grown in 4-inch pots and selected for use before blossoming. Leaves other than the 5 to be inoculated were trimmed away.

Inoculation of all plants was performed by rubbing each leaf with wooden pot-labels moistened with juice obtained by crushing infected tissues between two of the labels. During inoculation each leaf was sup-

ported on one, while its upper surface was rubbed with the other, of the pair of inoculum-moistened labels. A small quantity of 320-mesh carborundum powder was added to the inoculum immediately before its use. In most cases, a few drops of water were also added to aid in spreading the inoculum. The pot labels used in each inoculation were then used to record the procedure; residual virus was thus effectively buried in the soil surrounding the inoculated plants. The hands ordinarily did not become contaminated with inoculum, and in no case did the hands touch experimental plants during the process of inoculation.

#### DERIVATION OF SYSTEMICALLY SPREADING SUBMUTANT STRAINS

Stocks of plants carrying the two localized mutant strains of tobacco-mosaic virus were observed for a period of about 3 weeks after inoculation before being discarded. At 10 to 12-day intervals new lots of plants were inoculated.

Plants in which symptoms appeared in young uninoculated leaves were used as possible sources of new variants, or submutants. Transfer was made directly from the obviously affected top leaves to healthy young tobacco plants. In all cases strains other than the originals were thus obtained.

In one series of observations, 136 Turkish tobacco plants were inoculated with each of the two source strains. With but few exceptions these inoculated plants showed one to a few characteristic lesions, averaging 5.8 lesions per plant for strain 1933B and 6.4 lesions per plant for strain 1940F in a series of 56 plants inoculated with each and observed especially to determine the incidence of localized disease after routine inoculation.

Variants appearing in 136 plants inoculated with strain 1933B were 20 in number; with one exception, they were of the same general type as the green-mottling distorting strain from which the mutant strain 1933B arose (Figure 1). The exception, variant 1933B4, was of a type rarely observed before. It produced in tobacco a spotty-green-mottling systemic disease. In this it closely resembled isolate No. 104 of Jensen (1937) and like this isolate it produced much smaller than normal lesions in *Nicotiana glutinosa*.

Invariably the submutant isolates were of much higher infectivity, as measured by inoculation of *Nicotiana glutinosa* plants, than were the stocks of the original nonsystemic mutant strains. This is clearly shown in the accompanying table for mutant 1933B and its submutant derivatives. The infectivity of the source strain in each case was so low that all lesions could be counted; in individual tests the numbers of lesions ranged from 0 to 21. The infectivity of derived isolates was so high that only approximate estimates of numbers of lesions were made; in individual tests these ranged from about 500 to more than 1400.

No variants appeared in the corresponding set of 136 plants inoculated with strain 1940F. This indicated a great difference in stability of strains 1933B and 1940F but did not mean that strain 1940F was incapable of mutating. In fact, in other sets of Turkish tobacco plants inoculated with this strain, systemic variants appeared occasionally and proved of substantially higher infectivity than the localized yellow-spot source strain,



1940F. With one exception (Figure 1) they were of yellow-mottling type ; the exception was a systemic necrotic-spotting strain, that has been studied in some detail as strain 1952D. This exceptional strain was of intermediate infectivity. It, in turn, occasionally gave derivatives of yellow-mottling types that appeared much like those obtained directly from the localized yellow-spot strain, 1940F.

The set of plants inoculated with strain 1940F served mainly as a control on methods of inoculating and maintaining the experimental plants. Supposed derivatives from the less stable strain 1933B conceivably might have been obtained as a result of contamination with strains of tobacco-mosaic virus during or before the process of inoculation or subsequently during maintenance of plants for observation. Assurance that they were not, was furnished by absence of derivatives of 1940F within the experiment. Inoculations in the two sets were alike in character. Inoculum for successive sets was always from local lesions in plants of the preceding set that had shown no evidence of the presence of mutants. In no case did a mutant appear in any plant after use as a source of inoculum and before its discard. Sets were maintained side by side in the greenhouse. Yet invariably mutants appeared in the one and not in the other set of inoculated plants, showing the very unequal tendency to mutation of the two otherwise comparable original nonsystemic mutant strains. After the experiment, stock cultures were maintained for 12 months with transfers every 10 to 12 days ; with one exception every transfer of strain 1933B (in each case to 16 plants of Turkish tobacco) gave rise to one or more green-mottling systemically-moving variants, yet no one of the corresponding transfers of strain 1940F gave rise to any detectable variant.

The 20 variants derived from 1933B were re-examined in groups by inoculating plants with suspensions produced from dried samples of infected leaves. A series of 5 young Turkish tobacco plants was used for each sample and a similar series of 5 plants was inoculated with the green-mottling *distorting* virus from which strain 1933B was originally obtained. Minor differences were detected between some members of the final series of derivatives and the original strain from which localized source strain 1933B had been derived. The differences were especially in intensity of chlorosis in mottled areas, degree of puckering in young affected leaves at onset of systemic disease, and a tendency to show flecks of necrosis in old affected leaves. With the exception of 1933B4, however, all clearly belonged to the green-mottling group of strains of tobacco-mosaic virus (Table 1). Even isolate 1933B4 might be considered to belong within or near this group, though it had more pronounced differential characteristics than the other derivatives listed in the table.

It was obvious that variants derived from strain 1933B did not as a group constitute reversions to the original type in the strictest sense, since some of them could be distinguished definitely from each other and from the original source virus. Whether all could be distinguished from each other on further study, as Johnson and Valleau (1946) found to be the case for 54 isolates from field collections, is not known. Reversion to a general type or class of strains, that characterized by systemic green-mottling and moderate distortion of leaves in the case of derivatives from 1933B, seems

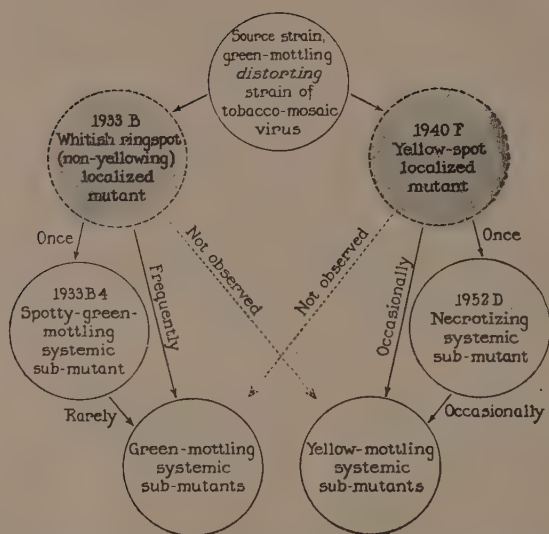


Fig. 1. Degree of infectivity correlated with ability of strains of tobacco-mosaic virus to move systemically in tobacco. Strains of low infectivity are indicated by dotted line circles; strains characterized by lack of systemic movement by stippling. Dependence of apparent infectivity and systemic movement is contrasted with independence of the characteristics of tissue-yellowing and other observed changes.



TABLE 1

*Character of derivatives from strain 1933B and comparative infectivity\* of derived strains and their source*

Designation of derivative	Character of derivative	Infectivity* of source strain	Infectivity* of derivative
1933B1	Yellow-green mottling	0	> 1250
1933B2	Green mottling	3	> 1100
1933B3	„ „	21	> 1200
1933B4	Chlorotic spotty mottling	5	> 1300
1933B5	Yellow-green mottling	6	> 1200
1933B6	Green mottling	13	> 1400
1933B7	„ „	2	> 1100
1933B8	„ „	2	> 650
1933B9	Yellow-green mottling	5	> 950
1933B10	Green mottling	1	> 650
1933B11	„ „	6	> 1350
1933B12	„ „	5	> 1150
1933B13	„ „	4	> 800
1933B14	„ „	2	> 1050
1933B15	„ „	3	> 1000
1933B16	Green mottling and etching	1	> 500
1933B17	Green mottling	6	> 1300
1933B18	„ „	8	> 600
1933B19	„ „	5	> 1300
1933B20	Yellow-green mottling	21	> 1150

\* Infectivity is expressed in terms of the number of lesions produced on 5 leaves of *Nicotiana glutinosa* within 5 days of subinoculation from tissues at the site of original inoculation, performed about 21 days earlier, in leaves of *Nicotiana tabacum*.

to have occurred in the present study rather than reversion to an exact model duplicating the original virus. Nothing even superficially resembling complete reversion occurred in mutation from 1940F, for no green-mottling strains at all were obtained from this source.

#### DISCUSSION

Derivation of submutants from nonsystemic mutant strains of virus has the technical advantage that separation from the original-source strain is essentially automatic. All submutants thus far observed have been capable of some movement to the uninoculated tops of infected plants. From affected top leaves these submutants have been recovered without admixture consistently, even by massive subinoculations to healthy plants.

The principal disadvantage of the process is the relative infrequency of appearance of systemic-type submutants. This disadvantage is offset by the improbability of obtaining a mixture of variants from any individual plant under these circumstances.

It will be noted that a confirmation of the earlier finding of independence between different mutations is presented in the series of experiments here reported and in related experiments with the same source strains. The schematic representation in Figure 1 shows that the original mutations provided characters that might be called *yellow-spot vs. nonyellow i.e., green spot*, in the two localized strains. These *yellow vs. green* characteristics recurred unchanged in observed systemic-type submutants, except in the case in which a necrotizing characteristic prevented recognition of such qualities in the systemic necrotic-spotting-type submutant 1952D, which subsequently, however, yielded yellow-mottling systemic variants. In other words, a factor determining yellow as opposed to yellow-green chlorotic effects, after appearing in one source strain of the present experiments, was represented throughout its own series of submutants, so far as could be determined, but was not observed at all in the series of submutants derived from the other source strain. The opposing tendency, to give rise to green-mottling submutants, was not observed at all during maintenance of the yellow-spot source strain, as indicated in Figure 1.

In contrast to this is the further observation that dependent or closely correlated changes occurred. That is, *all* submutants capable of moving to uninoculated leaves at the developing tops of inoculated plants were of much higher apparent infectivity than were the localized source strains. This raises the question whether the two apparently unrelated properties of a virus, infectivity and ability to move systemically, are dependent on a single mechanism within or on the surface of the rod-like viral particle or on separate mechanisms so related to each other spatially or chemically that a change in one strongly tends to involve a change in the other or in the effectiveness of the other. No exception to strict correlation has been noted thus far either during the change to localization or in the change away from it, whether in the present study, in previous experiments by the writer, or in experiments reported by others (Holmes, 1936 ; Jensen 1933, 1937 ; Kunkel, 1940, 1947).

In this simultaneous ability of a virus to undergo uncorrelated mutations and dependent mutations, the fundamental similarity between viruses and higher organisms is emphasized.

The physical difference between rod-like units of tobacco-mosaic virus, which can be freed from almost all surrounding materials without destroying their viability, and chromosomes of higher organisms, which cannot, greatly favors an effort to investigate the localization of genetic function by study of the viral structure. In connection with a non-rod-like virus, the bacteriophage T2H, evidence has been gathered by Hershey (1946) that independent factors exist as in tobacco-mosaic virus, and evidence for the view that linkage groups of mutational units exist in this bacteriophage has been presented by Hershey and Rotman (1948).

Whether apparently complete correlation of degree of infectivity and ability to move systemically in tobacco-mosaic virus should be viewed as a result of possible close linkage of gene-like structures within tobacco-mosaic virus is by no means clear. It might be explainable on a simpler basis if nonsystemic strains of virus should possess some extraordinary modification of form or an unusual tendency to aggregation, so that infective units would be large and few.

The independent and dependent mutations thus far demonstrated in tobacco-mosaic virus offer an opportunity to determine the nature of some mutational changes if chemical differences such as have been noted by Knight (1947) as occurring especially among natural variants of tobacco-mosaic virus can be distinguished and exactly correlated with the observed experimental mutations. No one has yet demonstrated controlled production of predictable types of mutation, as by chemical means, yet such control of mutation might become feasible if the actual difference underlying a particular type of mutation could be determined.

#### SUMMARY

It has been shown formerly that independent mutations are possible within tobacco-mosaic virus and indeed are common, as in higher organisms. Evidence is presented in the present paper to support the view that certain characteristics of this virus, namely infectivity and ability to spread systemically in infected tobacco, are either closely linked or actually controlled by a single mechanism, since they have been observed to mutate only simultaneously and not independently.

The Laboratories of The Rockefeller Institute  
for Medical Research, New York, N.Y., U.S.A.



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## GLUME BLOTCH OF WHEAT IN INDIA

B. L. CHONA

and

R. L. MUNJAL

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Glume Blotch of wheat caused by *Septoria nodorum* Berk., is well-known having been reported from Europe, America, and Australia but has hitherto not been described from India. During a recent visit to Kotagiri (Nilgiris, South India) in December, 1951, the disease was observed to be doing severe damage to local *Samba* wheat variety (*Triticum diccicum*) greatly affecting the seed setting and proper development of grain. Hardly 5 percent of the plants produced fertile ears and as a result thereof the whole crop had to be put for grazing. Luckily the area under this variety which was growing as a summer wheat, appears to be small and the infection was not found on other *Samba* varieties that were being grown as winter crop. It is necessary, however, to keep a careful watch in the locality in view of the serious potentialities of the disease. In India, another *Septoria* disease of wheat, Speckled Leaf Blotch caused by *Septoria tritici* Desm., is known to cause serious losses especially in Northern India but it chiefly affects the leaves and not the glumes as in the present case. The symptoms of the Glume Blotch disease and its casual organism are being described in some detail to facilitate identification of the disease both in the field and in the laboratory.

*Symptoms* :—As the name suggests, the disease manifests itself on the glumes as purplish-brown to brown, elliptic or oval spots varying from 4 to 9 m.m. in length and 2-3.5 m.m. in breadth and are surrounded sometimes by a yellowish halo. Later the spots turn greyish or straw coloured and are studded with black dots, the pycnidial fructifications (Fig. 1). The pycnidia are scattered singly but sometimes appear confluent due to their gregarious growth. In severe infection, it is not uncommon to find 2 or 3 spots on the glumes enveloping the entire surface of spikelets. Infection may extend even to the awns forming elliptic, brownish lesions which later become studded with black minute pycnidia, arranged irregularly in the diseased tissue (Fig. 1). Leaves, leaf-sheaths and rachis are also sometimes attacked but such infections are rather rare. On leaves and leaf-sheaths the spots formed, are yellowish to straw coloured and the affected portions dry up rapidly but on rachis the infection is similar to that on the ears. In contrast to this, the Leaf Blotch (*Septoria tritici*) remains mostly confined to the leaves, appearing as oval to elliptic spots, sometimes with well-defined margins. Several spots may be found on the same leaf and may cause the whole leaf to wither and dry up.

Perithecia resembling those of *Leptosphaeria* were also found on dried up leaves and leaf-sheaths affected with *S. nodorum*, intermixed with pycnidia, from which these were indistinguishable in a general observation.

*Causal organism* :—*Septoria nodorum* Berkley in Gard. Chron., p. 601, 1845.

Syn.—*Septoria glumarum* Pass.

*Macrophoma hennbergi* (Kuhn) Ber. & Vog.

*Phoma hennbergi* Kuhn

Histopathological studies of the affected tissues show the branched, hyaline, inter and intra-cellular mycelium ramifying the cells. The mycelium later collects in the substomal cavity and becomes faintly yellowish in colour due to the aggregation of protoplasmic mass. The sporogenous hyphæ give rise to pycnidia. Usually a single pycnidium is formed in one stromatic cavity but rarely two may come up together. Sometimes they are formed so close to each other that, when mature, they appear to be linear or gragarious. At this stage distortion of parenchymatous host cells commonly occurs. On leaves, the pycnidia are formed on both the surfaces but on the glumes, they are formed only on the outer surface. *Pycnidia* (Fig. 2) are globose or subglobose, subepidermal, later innate erumpent, yellowish brown in the beginning, later dark brown with a circular ostiole, about  $35\ \mu$  in diameter in the centre, and measure  $31.5\text{--}170\ \mu$  diameter, mostly  $112\text{--}160\ \mu$ . Cells around the ostiole are slightly smaller and darker in colour than those of the pycnidial wall. The cells of the pycnidium are parenchymatous in texture. Sections of the pycnidium show the wall to be 2-3 layers in thickness, the inner most layer supporting the pycnospores. *Pycnospores* come out in cirrhi from the pycnidium as in *Phyllosticta*. *Pycnospores* (Fig. 2) are hyaline, elliptic or cylindric, rarely curved, 1-3 septate, with upper end usually round and the lower slightly tapering, and measure  $14\text{--}26\ \mu \times 3.5\ \mu$ , mostly  $21 \times 3\ \mu$ . In *Septoria tritici* Desm. the spores are thinner, much bigger in length and slightly more curved. *Pycnidia* are also smaller in size.

*Ascigerous stage* :—As stated earlier under symptoms, perithecia were also observed intermixed with pycnidia in dried up leaves and leaf-sheaths affected with *S. nodorum*. Voglino (1904) discovered certain perithecia in the cultures of *Septoria nodorum*, which he identified as *Sphaerella exitialis* Morini. Weber (1922), who made detailed pathological studies on this fungus, found perithecia of *Leptosphaeria* associated with it and considered that Voglino's cultures were immature. Description of perithecia of our specimen which is presented below, agrees to a large extent with the recorded description of *Leptosphaeria* of Weber.

Perithecia are found embedded singly under the epidermis with the ostiole just coming out of the guard cells of stomata. They are globose or subglobose,  $85\text{--}120\ \mu$ , mostly  $105\text{--}112.5\ \mu$ , in diameter; parenchymatous, thin walled with broad, circular or oval ostiole measuring  $28\text{--}35\ \mu$ . Asci (Fig. 3) are clavate, numerous  $45.5\text{--}60\ \mu \times 5.8\ \mu$  with lower end slightly truncated, hyaline, thin walled and bear 8 ascospores. Ascospores (Fig. 3) are hyaline when immature but yellowish olive when mature, 3 septate, spindle shaped with both ends rounded, constricted at the septum, second cell somewhat bigger than other three cells, and measure  $15\text{--}20\ \mu \times 3.4\ \mu$ , mostly  $17.5 \times 3\ \mu$ . Paraphyses are simple, cylindrical, somewhat curved and multiseptate, hyaline, marginal, few and measure  $60\text{--}70\ \mu \times 3\ \mu$ .



*Artificial culture* :—Pycnosporos and ascospores germinated readily in tap water. They started putting forth germ tubes after about 4 hours' time at room temperature ranging from 21—25°C. Optimum germination was obtained at 20-22°C but there was no germination at 12°C or below, or at 30°C. Single-pycnosporos cultures on Potato dextrose agar and Oat meal agar produced innumerable black pycnidia after about 10 days growth with pink ooze of spore mass coming out of the ostiole. These resembled the original pycnosporos collected from the host in all respects except that these were mostly 2-3 septate. In single-ascospore cultural however, out of the 20 under study one developed typical pycnidia of *Septoria nodorum* thus indicating the possibility that *Leptosphaeria* sp. described above is the perfect stage of *S. nodorum*. Further cultural studies as also the cross inoculation tests are being carried out of confirm the above findings.

Grateful thanks are due to Dr. R. S. Vasudeva, Head of the Division of Mycology and Plant Pathology, I. A. R. I., New Delhi for his keen interest and helpful criticism in this work. Thanks are also due to Shri T. S. Ramakrishnan, Govt. Mycologist, Madras for the kind help in the collection of these specimens.

Division of Mycology and Plant Pathology,  
Indian Agricultural Research Institute,  
New Delhi.

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#### EXPLANATION OF PLATES

- Fig. 1. Wheat ears showing blotches and pycnidia on the glumes ( $\times 1\frac{1}{2}$ ) and a single affected spikelet further enlarged ( $\times 3$ ).
- Fig. 2. Pycnidium and Pycnosporos ( $\times 660$ ).
- Fig. 3. Ascus and Ascospores (Germinating) ( $\times 600$ ).



Fig. 1

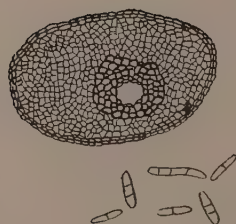


Fig. 2



Fig. 3

## A NEW BACTERIAL LEAF-SPOT AND STEM CANKER OF PIGEON PEA

Y.S. KULKARNI, M.K. PATEL

and

S.G. ABHYANKAR

(Accepted for publication April 20, 1952)

### INTRODUCTION

Bacterial leaf-spot and stem canker of pigeon pea, already described by the authors<sup>1</sup> is commonly found in Bombay during the months of July to September when the relative humidity is 80-90 per cent, air temperature ranges from 24°C. to 31°C. and the crop is in its early growing stage. Moreover, pigeon pea soon gets shaded by the rapid growth of sorghum which is grown as a mixed crop. All these factors, namely, shade, high humidity, air temperature and succulent stage of the host are conducive for the disease development. This paper gives a more extended account of the disease and its causal agent.

### DESCRIPTION OF THE DISEASE

Under ideal conditions, the pathogen produces small (0.5 mm.), round, water-soaked spots on the leaves after an incubation period of 7 days. The spots as they develop become quadrilateral (1 mm.) and are surrounded by a halo (0.5 mm.) on the upper surface of leaves (Plate I). Spots which are light-brown initially become dark brown later and are raised on the upper surface as a result of drying of bacterial exudation. The spots when coalescent form large lesions (2 mm.). In severe cases of infection, spots are found all over the leaf; the infection which extends to the main and lateral veins of the leaf, leaf-edges and the petioles results in a general yellowing of the leaf, cracking of the petiole and ultimate shedding. On the main stem and side branches, the pathogen produces dark-brown cankers (12.5 mm. long and 2.5 mm. broad) which when numerous and close cause peeling of the bark. A gummy bacterial exudate is also found by the side of these cankers.

### MORPHOLOGICAL, CULTURAL AND PHYSIOLOGICAL CHARACTERS

The organism is a short rod, Gram-negative, motile by a single polar flagellum, capsulated, readily stained with common dyes and measures  $1.3-2.2 \times 0.9-1.4$   $\mu$ .

On potato dextrose agar plates, colonies are smooth, shining with entire margins and pulvinate; colour is naphthalene yellow\*, diameter being 1.5 cm. after 7 days; colour of growth covering the entire surface of potato cylinders is straw yellow and of the cylinder light drab; in nutrient-dextrose broth, good cloudy growth in 24 hours; on nutrient agar plates, colonies round, shining, slightly raised, the colour being baryta yellow, diameter being 7 mm. after 4 days; on nutrient dextrose agar plates,

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1. Kulkarni, Y.S., M. K. Patel and S.G. Abhyankar (1950). A new bacterial leaf-spot and stem canker of pigeon pea. *Cur. Sci.* 19 : 384.

\*According to Ridgway's Colour Standards.



colonies round with entire margins, convex, 1.2 cm. in diameter after 4 days, colour being amber yellow ; milk peptonised ; litmus reduced ; gelatin liquefied, casein digested and starch hydrolysed ; acid but no gas in dextrose, lactose, maltose and sucrose ; salicin not utilised ; ammonia and hydrogen sulphide produced ; nitrates not reduced ; M.R. and V.P. tests negative ; sodium chloride tolerant up to 3 per cent ; non-lipolptic ; Loeffler's solidified blood serum liquefied in 10 days. No growth in inorganic basal medium containing separately each of the 10 amino acids viz. glycine, tyrosine, tryptophane, cystine, arginine, aspartic acid, glutamic acid, creatine, histidine, guanidine hydrochloride showing that the organism cannot derive its carbon from them. No growth in Koser's uric acid medium or in synthetic asparagin medium ; slight growth in Koser's liquid, and solid citrate media ; optimum temperature for growth 26-32°C. ; thermal death point about 51°C.

#### HOST RANGE

A wide host range including taxonomically distinct plants was included in the infection experiments. As usual, they were plants grown in earthen pots in the glasshouse one or two months prior to inoculation. The plants were kept in moist chambers before and after inoculation. The results conclusively showed that the pigeon pea varieties Nos. 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, F. 18 and F. 52 from Anand, M.4-N, M.4-E, M.4-5 from Arbhavi, Erect-C 47, Lonavala C-265, Matheran C-262, Kundal C-203, Kundal C-267 and Malawali comprising wild varieties were susceptible whereas *Arachis hypogaea* L., *Cyamopsis tetragonoloba* (L.) Taub., *Cicer arietinum* L., *Pisum sativum* L., *Stizolobium deeringianum* Bort., *Trigonella foenum-graecum* L., *Andropogon sorghum* Brot., *Oryza sativa* L., *Pennisetum typhoideum* Rich., *Ricinus communis* L., *Solanum melongena* L. and *Zea mays* L. showed no infection. Since the pathogen infects pigeon pea alone showing a host specificity, it is proposed as a new species to be called *Xanthomonas cajanii*.



Plate I

# STUDIES ON THE WILT DISEASE OF LENTIL (*LENS ESCULENTA* MOENCH)

R. S. VASUDEVA

and

K. V. SRINIVASAN

(Accepted for publication April 30, 1952)

## I. INTRODUCTORY

Lentil is a valuable pulse crop grown all over India, particularly in Bengal, Madhya Pradesh, Uttar Pradesh and the Punjab. Separate records of the acreage are not available for most of the states. The undivided Punjab, however, had an area of 215,000 acres producing an annual crop of 40,000 tons. It is a 'Rabi' crop, usually sown in October-November and harvested in March-April. The crop is grown in a variety of situations and on soils varying from the light loams to the heavy black cotton soils. A crop of the more humid regions of the U.P., it is grown as a "Sailaba" or inundation crop in the Punjab.

Records of the wilt of lentil are few. Prisyajnyuk (1931) reported a serious seedling blight due to a species of *Fusarium* in Russia. Fleischmann (1937) found lentil crops in Hungary, particularly in acid soils, liable to wilt but could not find any fungal or bacterial pathogen. Carrera and Noll (1941) found *F. avenaceum*, *F. scirpi* var. *acuminatum* and *F. culmorum* associated with wilts of lentil and blue lupin in Uruguay. In India mention of the disease is made in a report from Bengal (1934) where one of the imported varieties suffered badly from wilt. Padwick (1941) isolated a species of *Fusarium* distinct from *F. orthoceras* var. *ciceri* Padwick from wilted lentil plants. The wilt has also been reported to be caused by a species of *Fusarium* in the U.P. (Dey, 1948).

In February-March, 1948, wilt was observed in lentil N.P. type 1 in the Botanical area of this Institute. The disease was found to occur in a more severe form in 1949 and the incidence in some of the types under trial was observed to be as high as 67 per cent.

## II. SYMPTOMS OF THE DISEASES

In a broadcast crop wilt occurs in isolated patches more or less circular in outline which enlarge as the season advances. When the crop is sown in rows the disease appears to progress along the lines. Curling of the leaves begins from the lower end and extends upwards. The crown then droops and this is followed by the death of the plant. The root system is poorly developed and discoloured brown. The discolouration of the root system may be partial or whole. In early cases the tap root is destroyed at the tip and is abnormally short. Proliferation of secondary roots occurs in a cluster above the affected level. In a section of affected root the walls of some of the xylem vessels are discoloured brown and fungal hyphae are observed in some vessels, running close to the walls.

Observations made on the incidence of the disease in the field in early-sown crops show that some cases of wilt may occur in November and the disease almost ceases during the months of December and January. Again fresh cases of wilt appear in February and maximum incidence of disease is observed during March after which the incidence tends to fall in April. Fig. 1 shows the course of the disease as observed in two experimental plots on 16-10-48 and 15-11-48.

### III. EXPERIMENTAL

#### A. Isolation and pathogenicity tests

The causal organism was isolated from the affected roots in the usual manner and the fungus so isolated was purified by single spore technique. Large scale isolations made for this purpose yielded a species of *Fusarium*. Altogether 15 isolates were obtained and their pathogenicity tested. Soil in two pots was inoculated with each of the isolates and 6 seeds sown in each pot on 14th August 1948. Un-inoculated pots were kept as controls. Five isolates failed to bring about any infection whereas others showed infection varying from 9 to 33 per cent. The isolate which proved to be most virulent in these preliminary tests was selected for further investigation.

For infection tests relating to different experiments reported in this paper the fungus was multiplied on soil maize medium with the following composition :

Delhi soil (sieved through 1 mm. sieve) ...	152 gm.
Fine river sand ... ..	38 "
Finely ground maize meal ... ..	10 "
Distilled water ... ..	54 c.c.

The medium was put into 500 cc. Erlenmeyer flasks and autoclaved at 20 lb. pressure for two hours and inoculated. The flasks were kept at room temperature for 3 weeks. Sterilized Delhi soil of a loam type was used in all the pot-experiments.

Uniform quantity of the inoculum was placed about one inch below the seeds in pots. In the controls, soil-maize medium which had not been inoculated was used. Seeds were surface-sterilized for 2 minutes in 1 : 1000 mercuric chloride solution and washed in 3 changes of sterile distilled water before sowing. The experimental pots were kept in the room.

Lentil type N.P. 35 was throughout used in these experiments. Records were kept of the date of emergence and growth. Counts were made of the number of wilted plants. Re-isolations were usually made from the inoculated roots to confirm pathogenicity.

It was observed that seedlings emerged late, i.e. 3-7 days in pots the soil of which had been infected. Wilt appeared in 15-20 days or even a month after emergence.

#### B. Identification of the causal organism

The classification of Wollenweber and Reinking (1935) was followed for identification purposes. The growth of the fungus was studied on several



media prepared according to Wollenweber *et al* (1925) at 30°C. Rice was steamed thrice for one hour each at intervals of 24 hours. All agar media were autoclaved at 15 lbs. pressure for 20 minutes and the medium was poured into each of the petri dishes of 90 mm. diameter and inoculated from 3-week old cultures of the fungus growing on potato-dextrose agar. For comparison of the colour of the mycelium, stroma and the medium after 10 and 20 days growth, Ridgeway's colour standards (Ridgeway, 1912) were used. Spores were mounted on agar blocks and measured.

At the end of 20 days the aerial mycelium was short and dense on potato-dextrose agar, hard potato agar, P.D.A. with 5% dextrose, Brown's medium with starch and oat agar. It was more abundant on potato plugs and rice. On bean pods, *Melilotus* stems and lentil stems, there was a strong, coarse growth. The mycelium collapsed and formed a tough gelatinous mat after about 3 to 4 weeks on potato plugs, bean pods and rice. The characters regarding growth, colour of mycelium and the substrate as also the spore measurements of the fungus are given in Table I.

Stroma, thin plectenchymatous, not sclerotial-erumpent, pale to purple. Aerial mycelium abundant, wooly, white, sometimes turning pink, collapsing after 3 to 4 weeks and becoming gelatinous, tough, pionnotes and sporodochia absent. Conidia formed freely on mycelium at the ends of free conidiophores. Microconidia often agglutinated into false heads, non-septate, ellipsoid to avoid, cylindrical, oblong or slightly curved, hyaline. Macroconidia nearly straight to fusiform-falcate, slender with thin delicate walls and indistinct septa, conidia mostly single-celled, a few 1—5 septate spores present. Base papillate or tending to footcell formation, chlamydospores terminal or intercalary, borne on mycelium rarely on conidia, spherical to pyriform, smooth, hyaline, rich in protoplasmic contents, 1-celled, occasionally 2-celled, abundant on certain media, rare to absent on others. Abundance of ovoid to fusoid microconidia, delicate walls, and inconspicuous septation of macroconidia and occurrence of terminal and intercalary chlamydospores place it in the section *Elegans*. Absence of sporodochia further places it in the subsection *Orthocera*. The purple colour of the stroma, the colour and character of the aelial mycelium, and the measurements of the spores agree with the description of *Fusarium orthoceras* App. et Wr.

In pathogenicity tests twelve of each of the following plants were raised in infected soil. Six plants in sterilized soil served as controls in each case. None of the plants except lentil took infection.

Pigeon pea (*Cajanus cajan* (Linn.), Millsp.)  
Gram (*Cicer arietinum* L.)  
Broad bean (*Vicia faba* L.)  
Pea (*Pisum sativum* L.)  
Sweet-pea (*Lathyrus sativus* L.)  
Blue lupin (*Lupinus angustifolius* L.)  
Lentil (*Lens esculenta* Moench)

The lentil wilt organism is morphologically similar to the fundamental species *Fusarium orthoceras*. App. and Wr. It has a pale to purple stroma. Pionnotes, sporodochia and sclerotia are absent. Microconidia are abundant and macroconidia comparatively rare. It is not pathogenic to any of the hosts

TABLE  
Growth characters and

Medium	Colour of mycelium	Colour of growth on substrate	Colour of medium	0 Septate			1 Septate		
				%	Av. size $\mu$	Range $\mu$	%	Av. size $\mu$	Range $\mu$
P.D.A. 2%	White at first, later flesh pink in places	Pale to pansy purple	Pink turning violet carmine	83.7	8.5x 2.7	6.1- 12.1x 2.4- 3.4	6.7	17.2x 3.2	13.3- 21.7x 2.3- 3.8
Hard Potato Agar	White flecked with pink	Light perilla purple	Blackish brown	95.4	8.8x 2.7	6.1- 11.9x 2-3.4	2	17.5x 3.1	15.3- 20.4 x2.4- 3.4
P.D.A. 5%	White formaline Pink in Places	Cream	Purple	—	—	—	—	—	—
Potato plug	White, pink in places	—	—	96	7.9x 2.4	6.8- 11.9 x2-2.7	3	14.2x 2.6	11.9- 18.7x 2.4- 3.4
Brown's medium with starch	White at top, touched with pink in the middle	Slate purple	Perilla purple	76.3	8.1x 2.7	6.8- 11.9x 2.4- 3.4	8.7	19x 13.1	17- 23.8 x2.4- 3.4
Oat Agar	White	In places Bishop's purple	Not discoloured	—	—	—	—	—	—
Bean pods	White, turning pale	—	—	—	—	—	—	—	—
Meli-lotus stems	White, touched with olive buff in upper portions	—	—	93	8.4x 2.6	6.8- 10.2 x2-3.4	3	15.9x 3	11.9 18.7x 2.7- 3.4
Lentil stems	—	—	—	—	—	—	—	—	—
Rice	White, light buff on sides, later cream, places on top turning chatenay pink to flesh pink on addition of alkali violet	—	Dark perilla purple	95.9	9.1x 2.9	7.6- 11.0 x2.3- 3.8	1.1	15.2x 3.2	11-20.9 x2.7 3.8

## I

*size of the fungus*

2 Septate			3 Septate			4 Septate			5 Septate			Chlamydo spores	
%	Av. size $\mu$	Range $\mu$	%	Av. size $\mu$	Range $\mu$	%	Av. size $\mu$	Range $\mu$	%	Av. size $\mu$	Range $\mu$	Av. size $\mu$	Range $\mu$
5.4	22.6x 3.7	17.1- 24.7x 3.4- 3.8	4.6	34.6x 3.8	24.7- 47.9x 4-2.6	0.2	3-5.5 x4.1	—	Rare	44.5x 4.1	—	8.4x 6.5	6.8- 11.9 x5.9- 10.2
—	—	—	2.6	30.6x 3.2	27.2- 43.5x 2.7- 3.7	—	—	—	—	—	—	Few	6.8x 5.1
—	—	—	—	—	—	—	—	—	—	—	—	—	—
—	—	—	1	33.4x 3.6	23.8- 37.4x 2.7- 4.4	—	—	—	—	—	—	Abundant 0 Sep- tate 10.8x 8.7	6.8- 13.3 x6.8- 11.6
—	—	—	11.6	38.9 x3.7	33.3- 42.5x 3.4- 4.1	—	—	—	3.4	50.5x 3.8	42.5- 66.3x 3.4- 4.8	—	—
—	—	—	—	—	—	—	—	—	—	—	—	—	—
—	—	—	—	—	—	—	—	—	—	—	—	—	—
—	—	—	3	26.2 x3.1	23.8- 35.7x 2.7- 3.4	—	—	—	1	38.9 x3.5	37.4- 40.8x 3.4- 3.7	0 Sep- tate 9.7x 8.2	6.8- 13.3 x6.4- 11.9
—	—	—	—	—	—	—	—	—	—	—	—	—	—
—	—	—	3.1	34.4 x3.8	28.5- 41.8x 3.4- 4.8	5	spores	43.2x 3.9	—	—	—	0 Sep- tate 10.6x 8.9	6.4- 13.6 x6.4- 11.9



parasitised by the other varieties and in the present studies has infected only lentil. It is, therefore, proposed to name the fungus responsible for wilt disease of lentil as *Fusarium orthoceras* var. *lentis* var. nov. Vasudeva and Srinivasan.

Morphologically indistinguishable from the fundamental species. Cause of a vascular wilt of *Lens esculenta* Moench. in Delhi, India.

*Fusarium orthoceras* var. *Lentis* var. nov. Vasudeva and Srinivasan  
Organismum, quamquam morphologice distingui non potest a specie typica tamen nullas inficit plantas, quas aliae varietates infectant. Nova varietas producit marcescentism vascularem *Lentis esculentae* Moench. in urbe Delhi, India. Nova haec varietas hic proponitur, subnomine *Fusarium orthoceras* var. *lentis*, var. nova, Vasudeva & Srinivasan.

### C. Cultural studies

The fungus on Potato dextrose agar failed to grow at temperatures 9° and 35°C. whereas 27-30°C. was observed to be the optimum for growth. Also the fungus can tolerate a wide range of pH and that good growth is obtained between pH 4.6-5.8.

Effect of soil aeration on growth of the fungus was studied in glass columns 1" in diameter and 12" long. Greater the quantity of sand added, better is the aeration of soil. The different columns contained soil, soil-25% sand, soil-50% sand and pure sand, to each of which 1% finely ground maize had been previously added. The columns were plugged at both ends and sterilized. The columns were inoculated at one end with the fungus and kept at room temperature. Linear growth of the fungus along the wall of the tubes was measured on alternate days. The experiment was conducted in triplicates at 30°C. The growth of the fungus as recorded in different columns is given in Table II.

TABLE II

*Effect of soil aeration on the growth of the fungus*

Growth (days)	Average length of growth along the column in mm.			
	soil	75% soil 25% sand	50% soil 50% sand	Pure sand
4	4	6	7	10
6	11	15	17	22
8	18	24	28	37
10	25	33	39	52
12	32	42	50	68
14	40	52	60	80
16	47	61	70	92
18	53	69	80	105
20	61	78	91	117
22	67	85	103	131
24	74	92	113	146
26	79	99	125	159
28	84	107	126	168
30	89	116	148	174

The growth was most rapid on pure sand and was slowest on soil. The mixtures of sand and soil showed intermediate growth rates.

#### D. *Some factors influencing incidence of the disease*

(a) *Soil Moisture* :—The effect of soil moisture on wilt was determined in specially made metallic pots (Vasudeva, 1937) 7" high and 6" wide at the top with a small bent tube inserted at the side about half an inch above the bottom. This tube was used for irrigating the pots. The pots were filled with pebbles about 1/3rd inch in diameter just above the level of the irrigation inlet. Sieved soil which had been previously air-dried after sterilization was only used in this experiment. Twenty pots were filled with soil and divided into 4 lots. In each lot 3 pots were inoculated and 2 were kept as controls. Ten seeds of lentil were sown in each pot on the 1st November 1948. The moisture level in each lot was maintained at 25, 50 and 75 and 100 per cent of the water holding capacity by weighing the pots daily and adding sufficient water to make up the loss.

Germination was delayed in the inoculated pots maintained at 25 per cent soil moisture. Wilt appeared 15 days after germination in this lot, and 19 days after germination in the lot maintained at 50 per cent soil moisture. No plants, however, wilted in the inoculated pots maintained at higher soil moisture. Controls in each case remained healthy. The results are set out in Table III.

TABLE III

*Effect of soil moisture on lentil wilt*

Soil Moisture (Percent saturation)	No. of plants		Percent Wilt
	Inoculated	Wilted	
25	30	28	93.3
50	30	5	16.7
75	30	0	0
100	30	0	0

In order to see whether wilt would occur if the soil moisture of the pots maintained at 75 and 100 per cent soil moisture was reduced to 25 per cent, one pot in each of the treatments was brought to 25 per cent saturation after completion of observations in the foregoing experiment. In the case of pot originally maintained at 75 per cent saturation, wilt appeared 8 days after reducing the soil moisture and 6 plants died in the course of the week, whereas in the case of the pot originally maintained at full saturation wilt appeared 15 days after bringing down the soil moisture and 4 plants died as a result of wilt. In the corresponding controls in which the soil moisture was reduced accordingly, the plants remained healthy.

(b) *Soil temperature* :—In the absence of soil temperature control tanks, each lot of 10 pots containing inoculated plants and 5 pots containing un-inoculated plants to serve as checks were placed in 3 different places, *e.g.* in shade, in a glass cage and in open on 24-2-1942. Soil temperature in the pots placed in different places was recorded regularly. Soil moisture in all the cases was maintained at approximately 25 per cent saturation of the soil which is favourable for the development of wilt. Wilt appeared on 14-3-1949 and the maximum number of plants had wilted by 24-3-49. The results are summarised in Table IV.

TABLE IV

*Relation of soil temperature to wilt*

Treatment	Number of plants		Per cent wilt
	Inoculated	Wilted	
Shade (13-23°C.)	53	4	7.5
Cage (17-31°C.)	55	53	96.4
Open (13-38°C.)	48	20	41.7

Maximum number of plants wilted in the cage where the temperature varied from 17° to 31°C., whereas the disease incidence was lowest in the plants placed in the shade. The plants in un-inoculated control pots in each lot remained healthy.

(c) *Soil aeration* :—Effect of soil aeration on wilt was studied in metallic pots described previously. Mixtures of various proportions of well-sieved Delhi soil and river-sand were used. After thorough mixing, the different lots of sand and soil were sterilized separately and their water holding capacities determined. During the experimental period the soil moisture was maintained at 35 per cent saturation capacity in each case. Twenty-five pots were divided into 5 equal lots and each lot filled with soil and soil-sand mixtures. Three pots in each lot were inoculated and 2 pots were kept as un-inoculated controls. The seeds of lentil were planted in each pot on 27-2-49. Delay in germination and stunting of the plants were obvious in inoculated sand lot. The controls containing different mixtures showed little difference as regards germination and remained healthy. Wilt was first observed to appear on 24-3-49 in pots containing pure sand and 75 per cent sand, and 3 to 5 days later in the other treatments. The results of the experiment are set out in Table V.



TABLE V

*Effect of soil aeration on wilt*

Medium	No. of plants		Per cent wilt
	Inoculated	Wilted	
Soil ...	30	3	10.0
25% sand + 75% soil ...	30	4	13.3
50% sand + 50% soil ...	30	17	56.7
75% sand + 25% soil ...	30	27	90.0
Sand alone ...	30	30	100.0

The maximum wilt was observed in the lot raised on sand. The percentage of wilt fell as the quantity of soil increased in different lots, with the result that the lowest percentage of wilt occurred in soil alone.

Thanks are due to Rev. Dr. H. Santapau for providing Latin translation of the description of the fungus.

## SUMMARY

1. A new variety of *Fusarium orthoceras* proposed to be called *Fusarium orthoceras* var. *lentis* var. nov. Vasudeva & Srinivasan has been shown to be the cause of wilt disease of lentil.

2. A soil moisture of 25 per cent and periods when temperature ranged from 17°C-31°C. have been found to be most favourable for the disease. Also the causal organism attacks more vigorously when the plants are raised in sand as compared to soil.

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IND. AGR. RES. INST.,  
NEW DELHI

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# ALTERNARIA BLIGHT OF LINSEED

H.C. ARYA AND R. PRASADA

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## INTRODUCTION

India is one of the major linseed producing countries of the world. The crop which is raised mainly for extracting oil from the seed suffers from a blight disease which was first observed by Dey (1933) in January 1928, at Kanpur. He isolated the fungus but without making a detailed study of the organism, provisionally named it *Alternaria lini*, n.sp.

The disease is mainly confined to the floral region, as the affected flower buds are in most cases completely invaded by the fungous mycelium and its fructification. In such flowers grain formation or even the fertilization of the ovule does not take place. Such a disease has not been reported from any other country, although Badayeva (1930) observed spots on leaves and stems of flax caused by *Alternaria* in Siberia. Tervet (1937), Rost (1938) and Flor (1940) have reported that species of *Alternaria* were responsible for causing a seedling rot. Johnson (1942), and Ruschmann and Bartram (1944) have reported the occurrence of *Alternaria tenuis* Nees on flax straw which had been dew-wetted. Grove and Skolko (1944) isolated a species of *Alternaria* from the seed of flax and named it *A. linicola*. Moore (1946) also obtained *A. linicola* from flax seedlings in which the affected cotyledons had been partially or wholly blackened owing to the presence of numerous tufts of conidiophores and conidia; dark spots were observed on the first pair of leaves and pale brown streaks on the stems.

There was a severe outbreak of the 'blight' disease on the linseed crop of this Institute in March-April, 1949. In view of the seriousness of the disease and the confusion that exists in its correct identification, comparative morphological, physiological and pathological studies of the causal organism and allied species of *Alternaria* were undertaken, and the results are reported in this paper.

## SYMPTOMS OF THE DISEASE

The disease manifests itself in three sharply defined stages of floral development:

- (i) Before the fertilization of the ovule when the flower is in the bud stage,
- (ii) Just after the fertilization of the ovule, and
- (iii) After the maturation of the capsule (fruit).

In (i) the first indication of the disease is the failure of the buds to open during the day. Dark-brown spots appear near the calyx, get



enlarged, deepen in colour and spread all over the bud, passing into the pedicel. The petals and other floral structures shrink and become completely covered with the mycelial growth and its fructifications. The pedicel also shrinks, resulting generally in the collapse of the entire flower bud [Plate 1, fig. 1 (1)].

In the second stage where the infection takes place just after the fertilization of the ovule, the capsule develops normally, but it is very much reduced in size. The entire capsule turns brown in colour showing minute dark spots. The sepals present a burnt appearance. In such cases either seed formation does not take place or the seed is extremely distorted and shrivelled [Plate I, fig. 2 (2)].

At the third stage, the infection remains confined to the outer layers of the capsule as the dark spots or the discolourations are present only on the capsule coat. In this case also, the sepals give a burnt appearance but, since by this time grain formation has already taken place, no appreciable damage is caused and the seed develops more or less normally.

When the leaves are infected, dark brown spots appear, first in small irregular patches but subsequently spreading and passing over to the stem. The leaves ultimately get dried up and curled.

Unlike the symptoms produced by *Alternaria linicola* as described by Moore (1946), cotyledons do not show any symptoms of infection.

#### MATERIAL AND METHODS

The fungus was isolated from the infected flower buds and incubated at 26°C. All the isolates yielded *Alternaria* which was purified by taking single spore cultures. Linseed variety N.P. 12 was used for inoculations on account of its extreme susceptibility to the disease in the field. *Alternaria brassicae* (Berk.) Sacc. from *Brassica oleracea*, *A. tenuis* Nees from wheat seed, and *A. solani* (Ell. & M.) Jones & Grout from potato obtained from Indian Type Culture Collection of Fungi of this Division were used for comparative study. In every case conidia from ten days old cultures were used. Inoculations were made either by placing the conidia directly on the flower buds and leaves of plants grown in pots with the help of lancet needles or by spraying the plants with the conidial suspension by means of an atomiser.

#### EXPERIMENTAL

(1) *Cultural Studies* : When the fungus is grown on Potato-dextrose agar, the mycelium is almost colourless, appearing greyish in mass, and sparsely septate. It does not form any web during growth. Hyphae are 2-5  $\mu$  thick with average diameter 3.5  $\mu$ . The conidiophores are olive brown in colour, more closely septate than the vegetative mycelium, specially at the ends, wherefrom conidia are formed. The conidia are nearly linear to obclavate in shape and borne on the conidiophores in chains. Some are irregularly globular in shape. The chains mostly consist of 4-6 conidia, produced abundantly in the cultures as well as on

the host. They are olivaceous to brown in colour, smooth-walled at first, becoming roughened with age. The conidia including the beak call, consist of one to eight cells, vertical septations are few and rare, mostly the second or third cell is vertically septate. Beak cell in certain cases is absent ; if present, it is unicelled rarely 2-3 celled. The conidia taper somewhat towards the apex and are slightly constricted at the septa. Secondary conidia are formed in abundance in the cultures.

A comparative account of the measurements of different species of *Alternaria* grown on Potato-dextrose agar is given in Table I.

TABLE I.

*Measurements of conidia of different species of Alternaria on Potato-dextrose agar grown at 30°C.*

Fungus	Conidial formation	Measurements
<i>Alternaria lini</i>	Abundant	10.2-40.5 $\times$ 5.0-13.5 $\mu$
<i>Alternaria brassicae</i>	Abundant	10.0-40.4 $\times$ 5.0-14.0 $\mu$
<i>Alternaria solani</i>	Abundant	14.0-45.0 $\times$ 5.0-17.0 $\mu$
<i>Alternaria tenuis</i>	Sparse	14.0-30.0 $\times$ 5.0-12.0 $\mu$

Conidial size, therefore, shows a close similarity between *Alternaria lini* and *A. brassicae*.

In cultural characters, *Alternaria tenuis* differs from all others in forming dark grey coloured colonies with abundant aerial mycelial growth. Also, conidia and conidiophores are not so abundantly formed by *A. tenuis*. *Alternaria solani* can be distinguished from the rest by the presence of a long septate beak in the conidia and the dark black colour of its colony which is not differentiated into zones. *Alternaria lini* and *A. brassicae* resemble each other in all the cultural characters, as well as in the size and shape of their conidia.

Temperatures between 10° and 35°C. have been found to be suitable for growth with optimum at 26°-30°C. No growth was observed at 4° and 42°C. *Alternaria tenuis* is comparatively a fast growing fungus and *A. solani* a slow one, whereas there is no appreciable difference in the growth of *A. lini* and *A. brassicae*. All the fungi under study can withstand a wide range of pH (3-8.5) but the optimum lies between 5-6.5.

(2) *Pathogenicity tests* : Pathogenicity of the fungus was tested at different stages of growth by the following methods :

- i. Seed infection
- ii. Seedling inoculation
- iii. Inoculations of leaves and flowers of adult plants

For seed infection, seed obtained from apparently healthy capsules was surface sterilized by dipping it in 0.1 per cent. mercuric chloride solution for one minute and then washed thoroughly with sterilized water.

The seed was then inoculated by placing conidia on the seed coat and sown in pots containing sterilized soil. Equal number of un-inoculated seed was sown in another set of pots to serve as control. It was found that the germination of inoculated seed was only 18.7 per cent. as against 85 per cent. in the control. Examination of the ungerminated seed amongst the inoculated series revealed rotting of the radicles and plumules due to fungal attack.

Leaves of seedlings, and leaves and flowers of adult plants were inoculated by placing conidia on them with a blunt lancet needle, and incubating in a moist chamber for 48 hours. Successful infection was obtained after 72 hours and typical symptoms of the disease were produced.

Inoculations made at different times during the growing season resulted in cent per cent infection during January when the temperature range was 10°-28°C. and the incubation period was only three days. During December at 4°-23°C., 21.5 per cent infection was obtained and the incubation period was five days. During April and May when the prevailing temperature was between 27° and 40°C. there was no infection. The results are presented in Table II.

TABLE II.

*Results of inoculation tests conducted on different dates*

Date of Inoculation	Temperature (°C.)		No. of buds		Percentage Infection
	Minimum	Maximum	Inoculated	Infected	
16th December	4	24	70	15	21.5
23rd December	5	21	70	19	27.5
12th January	10	33	33	33	100.0
22nd January	7	31	50	35	70.0
14th March	24	36	30	20	66.6
13th April	26	39	30	0	0
1st May	28	40	30	0	0



Subsequent to inoculation, humidity seems to play an important part in determining the intensity of infection. Under optimum conditions of temperature, exposure to saturated atmosphere for 72 hours gave cent per cent infection. Exposures for 48, 24, and 12 hours resulted in 75, 8 and 5 per cent infection, respectively. Plants exposed for 6 hours to high humidity did not develop infection.

It is observed that the germ tubes of the fungus enter the host either through stomata or by direct penetration.

3. *Host Range* : In order to determine the host range of the *Alternaria* affecting linseed, plants belonging to the families *Cruciferae* and *Solanaceae* were inoculated because of the morphological similarity of *Alternaria* found on them. *Brassica oleracea*, varieties *capitata*, *botrytis*, *calorapa* and *Brassica campestris* var. *sarson* were successfully infected with typical symptoms. No infection was produced on *Solanum tuberosum*, *Lycopersicum esculentum* or *Nicotiana rustica*. Out of *Alternaria brassiceae*, *A. solani* and *A. tenuis* only *A. brassiceae* was able to infect linseed and produced typical blight symptoms [Plate I, fig 1 (2)].

4. *Viability of the fungus* : In order to determine the viability of the fungus during the summer months after the crop had been harvested, infected plant material was exposed to different treatments. Material kept in the laboratory where the maximum temperature during May and June was 40°C. gave cent per cent germination when tested in July, whereas, the material buried in the soil at a depth of three inches had lost its viability. This indicates that the fungus is able to oversummer in Delhi area only if the material is stored in a room. Preliminary tests with artificially infected seed show that Agrosan GN 2.5 : 1000 and Phygon 1 : 1000 are effective in eliminating seed infection.

#### SUMMARY

Morphological, physiological and pathological studies conducted show that the fungus responsible for the blight of linseed and named *Alternaria lini* Dey is identical with *A. brassiceae* (Berk) Sacc.

Temperatures between 26°-33°C. and humid conditions are most favourable for the growth of the fungus and the infection of plants.

Conidia can survive the summer heat in the room at Delhi but are killed under field condition. It has been indicated that seed infection may be controlled by the use of seed-dressing fungicides.

We are grateful to Dr. R.S. Vasudeva, Head of this Division, for his help and advice during these investigations.

Dn. of Mycology & Plant Pathology,  
Ind. Agricultural Res. Inst.,  
New Delhi.

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## EXPLANATION OF PLATE

- Fig. I (1) Symptoms of infection caused by *Alternaria lini* Dey  
 (2) Symptoms of infection caused by *Alternaria brassicae* (Berk.) Sacc.  
 (3) Healthy shoot of a linseed plant
- Fig. 2 (1) Seeds from healthy capsules  
 (2) seeds from infected capsules



Fig. 1

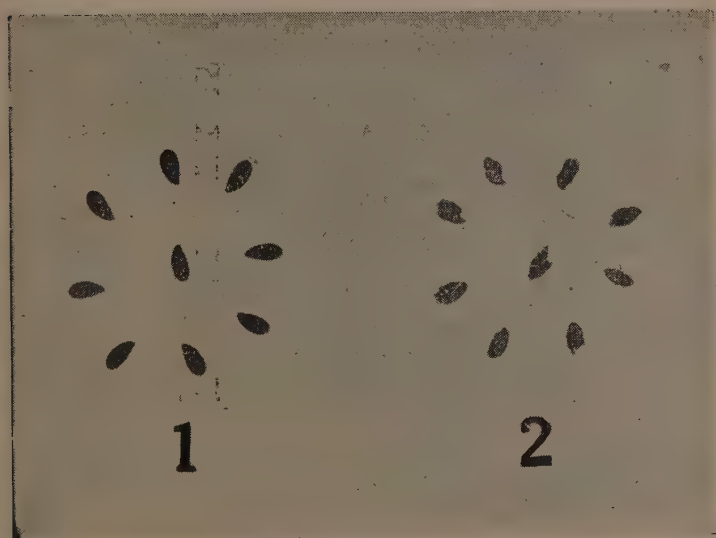


Fig. 2

## A COMPARATIVE STUDY OF ERWINIA CAROTOVORA, ERWINIA AROIDEAE, AND ERWINIA ATROSEPTICA

M. K. HINGORANI AND S.K. ADDY

(Accepted for publication May 5, 1952)

There has been considerable confusion in the identity and nomenclature of soft rot bacteria due to contradictory views put forth from time to time by various investigators. The literature pertaining to the subject has been reviewed by Stanley (1938), Waldee (1945) and recently by Rudd Jones (1950). An important question at the time, however, seems to be whether the three recognized species, viz. *Erwinia carotovora* (Jones) Holland, *Erwinia aroideae* (Townsend) Holland and *Erwinia atroseptica* (van Hall) Jennison, the black-leg pathogen, are synonymous or distinctive from one another. Leach (1930), Bonde (1939) and others consider that there is one species, *Erwinia carotovora*, and that *Erwinia atroseptica* with some six other named soft rot bacteria are synonyms of it, whereas Burkholder and Smith (1949) and Smith (1949, 1950) have separated *Erwinia atroseptica* from *Erwinia carotovora* on pathological and some physiological characteristics.

This paper presents a comparative study of five such isolates, including the authentic cultures of *Erwinia carotovora*, *Erwinia aroideae* and *Erwinia atroseptica*, obtained from Indian Type Culture Collection. The remaining two cultures were secured from decaying potatoes and onion bulbs.

### PATHOGENICITY

The isolates were first tested for their ability to infect potato tubers and cause a soft rot. The method used was that followed by Waldee (1945). Positive infection was obtained in all cases and typical soft rot developed after 24 hours of incubation period at 28°C. A jet black rot was always associated with *Erwinia atroseptica* which was easily distinguishable from the rots produced by the other cultures.

The isolates were also shown to cause soft rot of beet root, cabbage, carrot, cauliflower, cucumber, radish, knol kohl, onion bulbs, green peas and pepper, radish, turnip and tomato fruits. The type and colour of decay varied from culture to culture and from one host to another, but this character was not stable and was not of much value in separating the species.

Further inoculations were made on young potato plants of the variety *Phulwa* in the greenhouse to determine whether the isolates could produce the black-leg disease. The plants were inoculated when they were three to four inches high. Five plants were inoculated with each culture and proper controls were maintained. Inoculations were



made by injuring the stem at ground level and inserting the bacteria with a sterilized scalpel. The inoculated portion was then covered with sterilized moist cotton and the plants kept within bell jars for 48 hours. Successful infection occurred in the case of *Erwinia atroseptica* alone (Fig. 1).

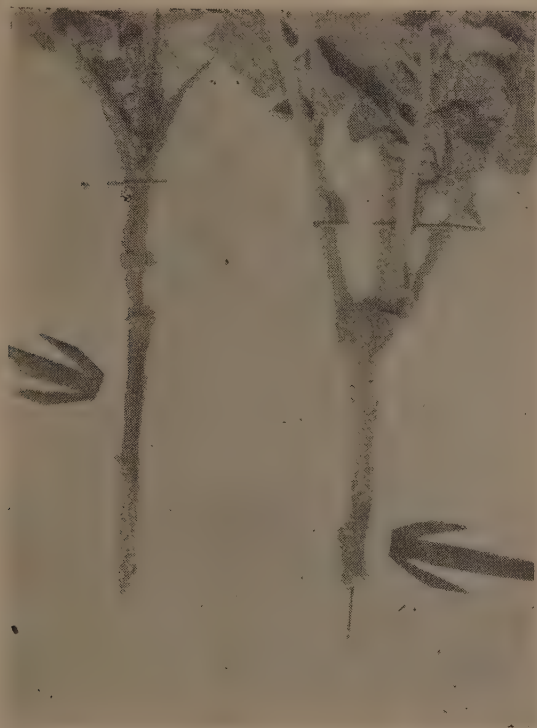


Fig. 1. Showing typical black-leg symptoms produced by *Erwinia atroseptica* on potato plants

Transmission of *Erwinia atroseptica* on a cutting knife and subsequent seed-piece infection was first suggested by Orton (1917), and, later on, experimentally proved by Smith (1950). Healthy tubers were, therefore, inoculated with the bacterial suspension of each of the five cultures by hypodermic injection method and incubated at 28°C. for 48 hours. Inoculated potatoes developed soft rot and a sterilized knife was used for cutting these tubers. After cutting a rotted potato with this knife, three healthy seed potatoes were cut in succession. A seed piece was cut with a sterilized knife to serve as control. The rotted seed-pieces cut with a sterilized knife, those inoculated with the cutting knife, and the controls were sown in sterilized soil in 4" pots and kept in a glass cage. It was found that, with *Erwinia atroseptica* infection, the transmission of the pathogen takes place by cutting knife resulting in missing plants and also in plants with black-leg symptoms. Other soft rot bacteria were not transmitted in this manner. Controls also did not show any infection,

## MORPHOLOGY AND PHYSIOLOGY

All isolates were similar in their morphological and physiological characteristics except in the utilization of certain carbon compounds. A summary of the fermentation indexes for the five cultures on six carbohydrate media is given in Table I.

TABLE I  
*The reaction of five soft rot bacteria on six carbohydrates*

Medium	Fermentation indexes*				
	<i>Erwinia atroseptica</i>	<i>Erwinia carotovora</i>	<i>Erwinia aroideae</i>	Potato isolate	Onion isolate
Glucose	0011	00?1	00?1	00?1	00?1
Sucrose	0222	0222	1111	0223	1122
Maltose	0222	0000	0000	0000	0000
Lactose	0222	0222	?111	0222	0112
Glycerol	0000	0011	00??	00?1	0001
Ethyl alcohol	0000	1111	0000	0111	0011

\* 0=no visible acid or gas ; ?=acid doubtful and no visible gas ;

1=acid but no visible gas ; 2=acid with less than 10% gas ;

3=acid with more than 10% gas.

First digit=reaction after 24 hours ; second digit=reaction after 48 hours ;  
third digit=reaction after 7 days ; fourth digit =reaction after 14 days.

It will be observed that *Erwinia atroseptica* utilizes maltose, producing acid and gas, whereas other cultures do not. It does not, however, assimilate ethyl alcohol and glycerol. *Erwinia aroideae* differs from the other cultures in not producing gas from any of the carbohydrate media. The two isolates from potato and onion correspond biochemically to *Erwinia carotovora*.

## CONCLUSION

The present investigation shows that various soft rot bacteria studied may be conveniently separated into two distinctive species, namely, *Erwinia carotovora* and *Erwinia atroseptica*. This has been done on the basis of differences in fermentation reactions and in pathogenicity. *Erwinia aroideae* is, however, considered here a non-gas-forming strain of *Erwinia carotovora*, thus confirming the findings of Dowson (1941).

The writers wish to express their thanks to Dr. R. S. Vasudeva, Head of the Division of Mycology and Plant Pathology, for helpful suggestions during the course of this investigation and for going through the manuscript.

Division of Mycology and Plant Pathology  
Indian Agricultural Research Institute  
New Delhi.

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## BLACK SPOTS OF MANGO FRUITS

B. N. UPPAL, M. K. PATEL AND M. N. KAMAT

(Accepted for publication May 10, 1952)

In 1937, Sir William J. Jenkins, then Director of Agriculture, during his tour of the coastal areas of Bombay State, came across mango fruits showing numerous black spots on their surface. These fruits were sent to us for diagnosis of the disease, and isolations made from such fruits gave a fungus which proved to be a new species of the monotypic genus *Actinodo-chium* first described by Sydow.\* The description of this fungus based on a detailed study of several isolations is given below.

### *Actinodo-chium Jenkinsii* sp. nov.

*Mycelium* — More or less effuse, septate, aerial, raised in close tufts, hyaline when young, turning smoke grey with age, growth best on potato dextrose agar with profuse sporulation, hyphae measuring  $3.5-10.5\ \mu$ . Minimum temperature for growth  $10^{\circ}\text{C}$ ., maximum being  $45^{\circ}\text{C}$ . and optimum  $30^{\circ}\text{C}$ ., resistant to freezing, grows best between 6 and 7 pH, although range lies between 2 and 9 pH.

*Conidia* — The hyphae break up into continuous long or short chains of short or long cylindric spores with rounded ends, subhyaline or grey, in chains of 2 to 20 or more (Plate I, fig. 2), measuring  $5-13\ \mu \times 3-5\ \mu$  with a mean of  $9 \times 4\ \mu$ . Germination by germ tube, minimum temperature for germination being  $10^{\circ}\text{C}$ ., maximum over  $40^{\circ}\text{C}$ . and optimum at  $26^{\circ}\text{C}$ .

*Chlamydospores* — More or less round, in chains, rarely single, walls smooth, rich in contents (Plate I, fig. 3) light olive grey to olive grey measuring  $6-14\ \mu$  with a mean of  $9\ \mu$ .

*Sporodochia* — Not found in nature or in cultures.

*Pathogenicity* — Pathogenic on fruits of alfonso and piari mango (*Mangifera indica* L.). Strictly (Plate I, fig. 1) a wound parasite. Ripe fruits are more susceptible than green ones. Causes few to several, dusky brown to blackish brown, round, necrotic spots measuring about 0.1 to 1 mm. Infection best at  $30-35^{\circ}\text{C}$ . Incubation period 72 hours. First collected at Vengurla by Sir W. J. Jenkins in 1937.

### *Actinodo-chium Jenkinsii* sp. nov.

*Mycelium* plus minusve effusum, septatum, aereum, elevatum in acervulis adpressis, hyalinum initio, fumosum ni maturitata; culturae medium optimum est Potato-dextrose-agarum, in quo est profusa sporulatio; hyphae magnit.  $3.5-105\ \mu$ .

\*Sydow, H. (1927) Fungi in itinere costaricensi collecti. *Ann. Mycol.* 25: 1-160



Conidia—hyphae dividuntur in breves longasve catenas sporarum longarum vel brevium, cylindricarum, rotundatarum in utroque apice, subhyalinarum vel grisearum, spora in singulis catenis 2-20 vel plures, magnitud.  $5-13\mu \times 3-5\mu$ , medietate  $9 \times 4\mu$ .

Chlamydosporae—plus minusve rotundae, catenatae, raro singulae, parietibus laevibus, materia in sporis contenta ampla; colore pallide ad plus minusve alte olivaceogriseae, magnit.  $6-14\mu$ , medietate  $9\mu$ .

Sporodochia—non visa in natura nec in vitro.

Pathogeneitas—In fructibus *Mangiferae indicae* Linn. var. "a[fonso" atque "piari"; stricte parasitus in fructuum vulneribus.

Culture deposited at the Commonwealth Mycological Institute, Kew, England.

Plant Pathological Lab.,  
College of Agriculture,  
Poona 5.

#### EXPLANATION OF FIGURES.

Figs. 1-3 *Actinodocheium jenkinsii*; 1. Natural infection spots on mango fruits. Fruit at the extreme right below is healthy; 2, Mycelium breaking up into chains of oblong conidia; 3. Mycelium forming chains of chlamydospores. Few oblong conidia are also present.

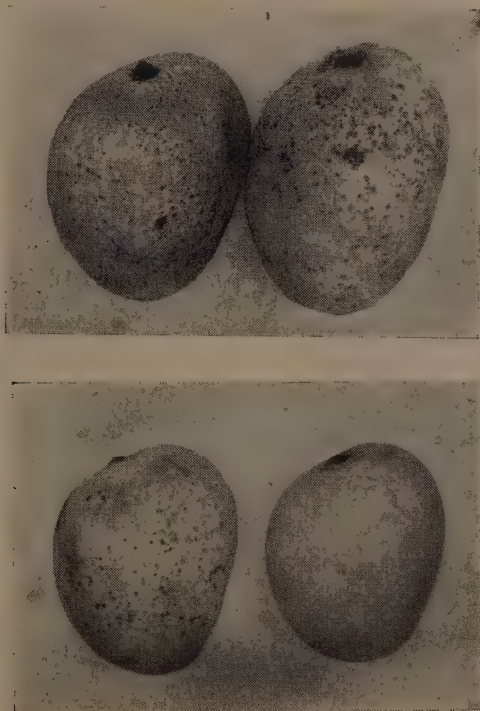


Fig. 1



Fig. 2

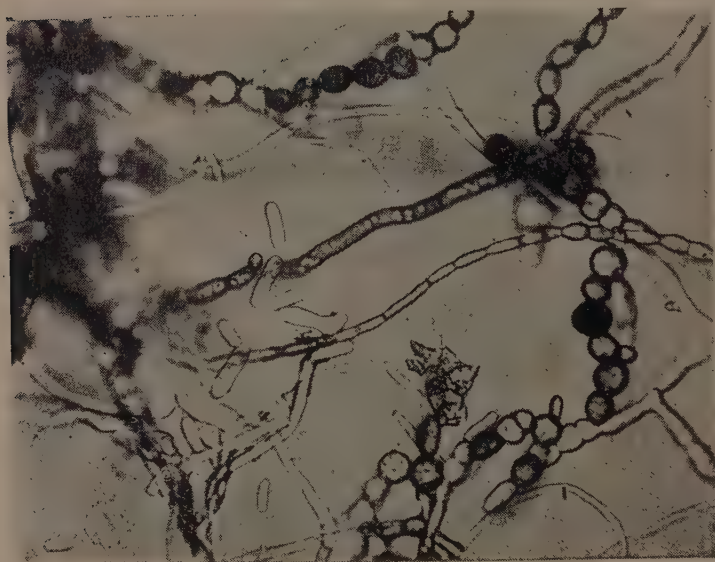


Fig. 3

## A MOSAIC DISEASE OF *NICOTIANA GLAUCA* R. GRAH.

T. K. NARIANI  
and  
NIRMALJIT SINGH

(Accepted for publication May 15, 1952)

In March 1951, a mosaic disease of *Nicotiana glauca* R. Grah. was observed in the crop of Botany Division of this Institute. About 20 per cent of the plants were found infected with the disease. In the following year the infection was as high as 40 per cent. The most prominent symptoms of the disease are the dark green mosaic mottling on the leaves and reduction in leaf size. The dark green areas are invariably raised and assume the shape of blisters, but at times they run parallel to the veins producing raised green vein banding effect (Plate I, Fig. 1). The leaves are slightly malformed and have wavy margins.

*Transmission* :— The disease was readily transmitted under glasshouse conditions by sap inoculation by the usual rubbing method and also by using carborundum powder as an abrasive. The artificially inoculated *Nicotiana glauca* plants develop vein clearing after about 15 days which is followed by mosaic mottling. The dark green mosaic symptoms develop at a later stage with slightly raised blister-like patches occasionally along both sides of the veins. There is malformation of the leaves accompanied by reduction in the leaf lamina.

*Host range* :— Inoculations with the sap expressed from the mosaic affected *N. glauca* plants were made on various members of *Solanaceae* and *Cucurbitaceae*. The disease was found to be transmissible to *Nicotiana tabacum* varieties *Harrison's Special* and *White Burley*, *N. glutinosa* L., *Lycopersicon esculentum* Mill., *Datura stramonium* L., *Capsicum annum* L., *Solanum nigrum* L., *S. nodiflorum* Jacq., *S. melongena* L., *Petunia hybrida* Vilm., *Cucumis sativus* L. and *Trichosanthes anguina* L. The virus however, could not be transmitted to *Luffa aegyptica* Mill., *Citrulus vulgaris* Schrad. *C. vulgaris* var. *fistulosus* and *Lagenaria leucantha* (Duch) Rusby. Potato varieties *Up-to-date*, *President*, *Arran Victory*, *Majestic* and seedling 41956 by sap inoculation did not take infection. The disease was however, transmitted by grafting to *Up-to-date* variety from which the virus was recovered back on tobacco. Varieties *President* and *Seedling* 41956 did not take infection even by grafting.

The main symptoms on tobacco varieties *Harrison's Special* and *White burley* are the pronounced vein clearing followed by pale green mosaic mottling with occasional blisters and green vein banding. *N. glutinosa* reacts in a characteristic way by producing severe blister type mosaic accompanied by reduction in leaf lamina which is reduced to almost filiform or tendril shape (Plate I, Fig. 2). On tomato the virus produces the typical "fern leaf" effect at low temperatures (Plate I, Fig. 3), but at high temperatures the plants carry the virus symptomlessly. Severe distortions, pucker-

ing and blistering accompanied by finger-like projections arising from the leaf margins are the chief symptoms on *Datura stramonium* (Plate I, Fig. 4). On *Cucumis sativus* minute greenish yellow angular chlorotic spots bounded by the veins appear and give a characteristic mosaic appearance to the leaves (Plate I, Fig. 5). Mosaic mottling is produced on *Capsicum anuum*, *S. nigrum*, *Petunia hybrida*, and *Trichosanthes anguina* while *S. nodiflorum* and *S. melongena* react by producing marginal mosaic and green vein banding mosaic respectively. The axillary shoots on the grafted *Up to-date* plants show mild mosaic symptoms accompanied by leaf curling and distortion.

*Properties* :— The properties of the virus were studied in the insect proof house with the infective juice obtained from the inoculated tobacco plants. The virus in the standard extract\* remains infective after an exposure to 57°C. for 10 minutes but it loses infectivity when exposed to 60°C. for the same period. Crude juice of the virus affected leaves when diluted to 1:100 with water was found to be infective, but at 1:200 dilution it was rendered innocuous. The virus in the standard extract was found to retain its infectivity after 7 days' storage at 6°-8°C., but was rendered innocuous after 8 days' storage at the same temperature.

A mosaic disease of *N. glauca* has been reported by McKinney (1929, 1944) from Canary islands. The virus produces local necrotic lesions on *N. glutinosa* and has a thermal death point of 86°C. *Lycopersicon esculentum* and *Cucumis sativus* are immune. The virus under study differs markedly from the above in physical properties, in its inability to induce necrotic lesions on *N. glutinosa* and ability to infect *Lycopersicon esculentum* and *Cucumis sativus*. The thermal death point and the reaction on cucumber, tomato, *Datura stramonium* and *Nicotiana glutinosa*, however, suggest a close resemblance with *Cucumis virus 1* (Smith, 1937). The "fern leaf" effect on tomato, typical mosaic symptoms on cucumber and *Datura* and blistering type of mosaic on *Nicotiana glutinosa* are the characteristic symptoms of *Cucumis virus 1*. The virus however differs in having a low dilution end point. The cause of the *Nicotiana glauca* mosaic disease may therefore be attributed to a virus which lies close to *Cucumis virus 1*.

Thanks are due to Dr. R. S. Vasudeva, Head of the Division of Mycology and Plant Pathology, for guidance and helpful suggestions throughout this investigation.

Division of Mycology & Plant Pathology  
Indian Agricultural Research Institute,  
New Delhi.

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\* 1 c. c. of distilled water added to the extract of every gram of infected leaf material.



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### EXPLANATION OF PLATES

#### Plate I

- Fig. 1. Mosaic affected leaves of *Nicotiana glauca*. Naturally infected.
- Fig. 2. *Nicotiana glutinosa* plant showing blister type mosaic and filiform leaves.
- Fig. 3. Tomato plant showing "Fern leaf" effect caused by the virus.
- Fig. 4. Infected *Datura stramonium* showing blistering and mosaic.
- Fig. 5. Leaf of infected cucumber showing typical mosaic mottling caused by the virus.



Fig. 1



Fig. 2



Fig. 3



Fig. 4



Fig. 5

## FUSARIUM ROSEUM LINK AND SCLEROTIUM ROLFSSII SACC. ON GINGER RHIZOMES

B. S. MEHROTRA

(Accepted for publication June 30, 1952)

Besides the well known soft-rot of ginger (*Zingiber officinale*) caused by *Pythium*, a number of other fungi have been reported, from time to time, as occurring on ginger rhizomes. Two fungi, viz., *Fusarium roseum* Link and *Sclerotium rolfsii* Sacc., isolated from ginger rhizomes, are here recorded.

### I. *Fusarium roseum* Link

So far *Fusarium* has never been reported on ginger except for a mere mention of an unidentified species of *Fusarium*, causing basal-rot, reported from Hawaii<sup>1</sup>. During the rainy season, in August 1951, the author observed stored ginger rhizomes covered with a white aerial growth of a fungus. Pure cultures of the fungus, obtained as usual and grown on Czapek's acid sucrose nitrate medium, showed the following morphological characters.

Sporodochia minute, pale to brown, conidia 0-5 septate, oval or long to spindle-sickle-shaped, mostly 3 and 5 septate but 5 septate more often. 0-septate,  $13-30 \times 2.5-3.3\mu$ ; 1-septate,  $16-33 \times 2.5-3.3\mu$ ; 2-septate,  $13-27 \times 2.5-3.3\mu$ ; 3-septate,  $16-40 \times 2.5-4.0\mu$ ; 4-septate,  $22-43 \times 2.5-4.0\mu$ ; 5-septate,  $30-50 \times 3.5-4.0\mu$  (Fig. 1).

Chlamydo-spores  $6-14\mu$  in diameter, round, smooth (smaller ones) or rough, on mycelia or in conidia, intercalary as well as terminal, more frequently many celled in chains or knots than one celled, younger ones hyaline, older in mass brown. (Fig. 1)

The fungus resembled in all respects with *Fusarium roseum* Link except that the size and septation of the conidia of this isolate varied from the varieties of *Fusarium roseum* Link so far described. Since measurements are of little value in *Fusarium* taxonomy, because of their great variability due to environmental and genetic factors, the author, in conformity with the suggestion of Dr. W. C. Snyder, has abstained from naming it a new variety.

The inoculation experiments on healthy rhizomes showed that the fungus is incapable of penetrating the healthy parts. Over the injured or decaying portions of the rhizome there was sufficient white aerial growth of the fungus. It is therefore concluded that the fungus is an occasional, secondary or wound invader of ginger rhizomes.

### II. *Sclerotium rolfsii* Sacc.

*Sclerotium rolfsii* Sacc. has a wide host range having been reported on a number of economic plants in India and abroad. In Ceylon, as early as

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<sup>1</sup>Anonymous (1938) Rep. Hawaii agric. Exp. Sta., 1937, 35-45.



1934, seed ginger stored at Paradeniya was found to be superficially infected with *Sclerotium rolfsii* Sacc<sup>2</sup>. In India it has never been reported so far and is here recorded for the first time.

In August, 1952, during the rains, some of the ginger rhizomes, in the local market, were seen having white patches of fungal growth. Pure cultures of the fungus on Czapek's acid sucrose nitrate medium and oat-meal agar showed the following morphological characters which resembled with those of *Sclerotium rolfsii* Sacc.

Mycelium densely floccose, bearing numerous, pinkish-buff to olive-brown to clove-brown, globose sclerotia, 0.8-2.5 mm. in diameter.

Further, this isolate was in all respects similar to a culture of *Sclerotium rolfsii* Sacc. maintained in the culture collection of Botany Department, University of Allahabad.

Inoculation experiments on healthy and sterilized ginger rhizomes showed that the fungus can grow only superficially and is incapable of penetrating the rhizome. There was at first a wooly growth of the fungus but later (after about a week) with the development of white to brownish sclerotia the mycelium almost disappeared. Further the fungus was also capable of growing from an infected rhizome to a healthy one (Fig. 2). The ginger rhizomes, except for the superficial growth of the fungus, remained apparently sound.

Thanks are due to Dr. W. C. Snyder for the specific identification of *Fusarium* and for his helpful suggestion.

Botany Department,  
University of Allahabad,  
Allahabad, India.

Fig. 1. *Fusarium roseum* Link  
1, Sporodochium ; 2-9, conidia ; 6, chlamydospore in conidium ; 10, terminal chlamydospores.

Fig. 2. *Sclerotium rolfsii* Sacc.  
Fungus from an originally infected rhizome (on the right) growing over an adjacent uninfected rhizome (on the left). The one on the right showing a number of sclerotia.

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<sup>2</sup>Park, M. (1937)—The seed treatment of ginger. Trop. Agriculturist, 89, 3-7.

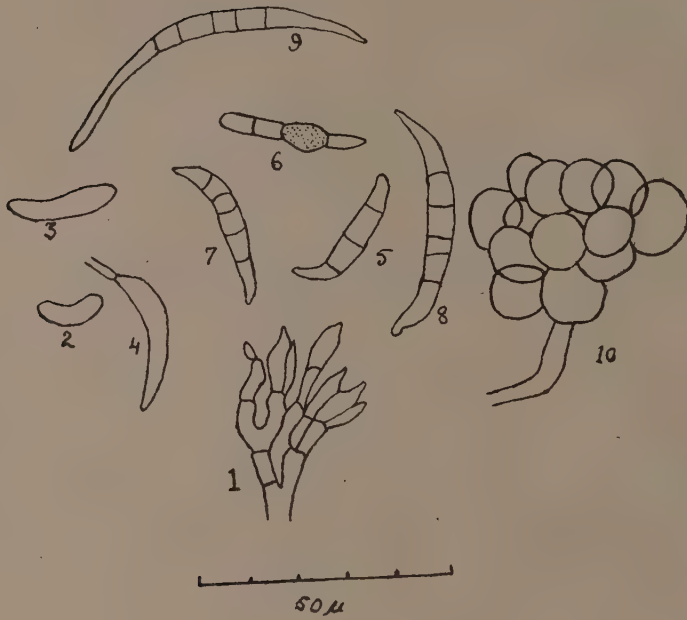


Fig. 1

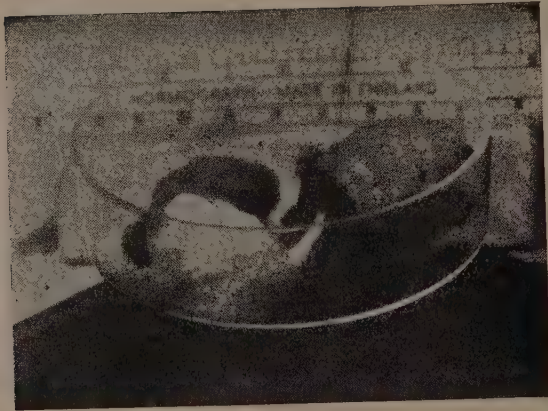


Fig. 2

## BACTERIAL LEAF SPOT OF POMEGRANATE

M. K. HINGORANI & P. P. MEHTA

(Accepted for publication July 30, 1952)

During the summer of 1951, a leaf spot of *Punica granatum* L. was observed in the orchard of Entomology Division. Irregular spots, varying from 2 mm. to 5 mm. in diameter, appear primarily on the leaves; but a number of adjacent spots may coalesce and cover larger areas. The initially light brown spots later turn dark brown and are surrounded by prominent water-soaked margin (Fig. 1). The formation of several spots on a leaf sometimes induces shedding.



Fig. 1

Microscopic examination of the lesions revealed the presence of myriads of bacteria. Dilution plates yielded the same organism every time. Infection occurred readily on the tender leaves of the young potted plants by spray method in the absence of wounds, the bacteria presumably entering through the stomata (Fig. 2). The incubation period was from 7 to 10 days, after which sections of the necrotic tissue showed large numbers of bacteria. Isolations from the infected plants yielded an organism similar to the one used in the inoculation experiments. Further tests showed they were identical.

The pathogen is briefly described in culture as follows. Short rods with rounded ends, single or in pairs, sometimes in chains, measuring  $1-2.5 \times 0.5$  microns in size; motile with a single polar flagellum, Gram-negative, no endospores, capsules absent, not acid-fast. Growth on agar plates is slow, filiform with edges entire, glistening, colourless to pale yellow, butyrous, and browning of the medium takes place at high temperatures. Nutrient broth turns cloudy within 48 hours; colonies on potato-dextrose agar are round, raised, and measure 1 to 2 mm. in diameter on the average;

profuse growth takes place in Clara's medium, but none in Uschnisky's solution. Stratiform liquifaction occurs in gelatin ; nitrites not formed ; ammonia, hydrogen sulfide and indole not produced ; starch not hydrolysed ; M. R. reaction positive ; V.P. test negative ; litmus not reduced but coagulation with subsequent peptonization takes place. Glycerol and salicin not utilized ; acid without gas produced in xylose, glucose, lactose, sucrose and raffinose. It is strictly aerobic. The optimum temperature for growth is about 30°C., maximum 40°C., and minimum about 5 - 10°C. The thermal death point is about 52°C.



Fig. 2

No bacterial pathogen has so far been described on pomegranate leaves. Further work on identification of the organism is in progress.

The writers wish to express their thanks to Dr. R. S. Vasudeva, Head of the Division of Mycology and Plant Pathology, for going through the manuscript.

Division of Mycology and Plant Pathology  
Indian Agricultural Research Institute  
New Delhi.



## FIFTH ANNUAL REPORT OF THE INDIAN PHYTOPATHOLOGICAL SOCIETY (1951)

I am submitting herewith the Fifth Annual Report for the year 1951 of the Indian Phytopathological Society. At the end of 1950 the total membership was stated to be 174. During 1951, four new members were enrolled. The total number of members in good standing now is 133 ; one is a Patron, 40 are Life Members, 92 have paid their subscription for 1951 and 5 have paid for 1952 also. 50 members have not paid their 1950 subscription so far in spite of repeated reminders and requests. It is earnestly hoped that they will pay up as soon as possible. In this connection I would like to point out that a considerable amount of time and money is wasted in sending reminders and I hope that the members will realise these difficulties and pay their dues on the first bill. Four members have expressed their intention to resign. They have been approached to reconsider their decisions and to continue their association with the Society.

Volume III, No. 1 and 2 of Indian Phytopathology were published this year. No. 1 has been posted to the members and the subscribers and No. 2 will be in their hands within a week. The Journal continues to enjoy great popularity in foreign countries and 25 new subscribers were added to the list of 82 this year bringing the total number to 107. This is very encouraging but unfortunately a large number of Institutions and Colleges are still not subscribing and individual members should exercise their influence to make their Institutions ask for the Journal if they are not already getting it.

As pointed out in my last report we had to change the press and entrust the work to a local press in the hope that we shall be better able to look after the printing work and expedite the issue of the Journal. Since the local press had not handled scientific material before, considerable difficulties were experienced in bringing out Vol. III in a satisfactory condition. Keeping in view the nature of work which the press had to handle, the output has not been bad and I crave the indulgence of the members and subscribers if some mistakes have crept in. Every possible care was taken and will continue to be taken to ensure the maximum amount of efficiency with the material at our disposal. Vol. IV, No. 1 is now in the galley proof stage and we shall make every endeavour to bring out Vols. IV and V before the end of 1952 thus making up for the arrears. To enable the Editor to fulfill this promise, the member should send their best articles for publication without delay because otherwise he can do nothing. Considering the wide circulation of our Journal it is no longer necessary for the research workers in this country to send their articles for publication abroad.

Putting aside a sum of Rs. 5,000/- invested in National Savings Certificates, the year began with Rs. 4,524/2/1 to our credit. Receipts during the year amounted to Rs. 4,016/12/- which includes Rs. 250/- given by the National Institute of Sciences of India as donation. The expenses incurred

during the year amounted to Rs. 1,989/9,3 only. This is due to the fact that no payments were made this year for the printing of the Journal. Next year we will have to pay for the printing of four issues, *i. e.* for Vols. III and IV, and may be for Vol. V also. Reckoning at the rate of Rs. 2,000/- as printing charges for each issue, a sum of Rs. 12,000 (Rupees Twelve Thousand) will have to be paid on this account alone during 1952. It is obvious that it will not be possible to meet this expenditure unless the membership and subscription fees are increased. It will also necessary to increase the number of members and subscribers. If every member were to undertake to enlist one more member and also makes an effort to see that his Institution becomes a subscriber, if not already so, it may still be possible to postpone the consideration of increasing the fee for another year in order to watch the results. I personally feel that we have reached a saturation point so far as the enrolment of members is concerned. We may at the most secure a few more members which will not solve the problem. A resolution is going to be put before you suggesting an increase in the fees which, I am sure, will receive your careful consideration. The cost of printing is gradually going up and there seems to be no alternative if we have to discharge our obligations efficiently. We have striven very hard to get the printing done as cheaply as possible and I am prepared to negotiate with any other press which will do our work at cheaper rates consistent with efficiency and standard.

The accounts for 1950 were audited by a Chartered Accountant at a cost of Rs. 25/- and are placed before you. They will be published in Vol. IV of the Journal. A tentative statement of Receipts and Expenditures for 1951 has been prepared and is submitted herewith. It will be properly audited by a Chartered Accountant as in the previous years.

Our grateful thanks are due to the Indian Council of Agricultural Research for subsidising the printing of the Journal, INDIAN PHYTOPATHOLOGY, by 33 per cent and to the National Institute of Sciences of India for the grant of Rs. 250/-.

I take this opportunity to express my grateful thanks to the members of the Society, the Councillors and Dr. M. K. Patel, our president for the support, encouragement and advice which I have received from them. My grateful thanks are also due to Dr. R. S. Vasudeva for giving me the necessary facilities and to Dr. B. B. Mundkur for helping me most ungrudgingly whenever I approached him.

Lastly, I would like to be excused for not presenting my report to you personally. I had to give up the idea of travelling to Calcutta for reasons of economy and am requesting my friend Dr. B. C. Chona to act on my behalf. I wish to thank him for this trouble.

R. PRASADA  
*Secretary-Treasurer*  
29 Dec. 1951

## THE INDIAN PHYTOPATHOLOGICAL SOCIETY, DELHI.

Receipts and payments account for the year ended 31st December, 1951.

RECEIPTS		PAYMENTS	
	Rs. A. P.	Rs. A. P.	Rs. A. P.
To Opening Balance :			
Cash in hand	185 8 3		1,277 1 0
Balance with Lloyds Bank			64 6 6
Ltd., New Delhi in Savings			400 0 0
Account	4,338 9 10		25 0 0
			85 0 0
Admission Fees			
Life Membership Fees			100 0 0
Subscription :			35 4 0
For 1949			37 8 0
" 1950	10 0 0		23 13 9
" 1951	103 0 0		
" 1952	870 0 0		
	46 10 0	77 3 0	
Subscription for Journal			
Reprints	1,863 5 9		
Advertisement in Journal	562 15 0		
Interest on Savings Account	216 0 0		
Publication Grants	39 8 0		
	250 0 0		
		6,498 4 7	6,575 7 7
	8,625 8 10		8,625 8 10

We have examined the above receipts and payments account and certify that it has been found correct in accordance with the books and information supplied.

We have also verified that Post Office Twelve-Year National Savings Certificates of the value of Rs. 5,000 belonging to the Society was held in Safe Custody with Lloyds Bank Ltd., as on 31-12-1951.

New Delhi :

R. PRASADA

Secretary-Treasurer.

AIYAR &amp; Co.

Chartered Accountants.



**Minutes of the Fifth Annual General Meeting of Indian Phytopathological Society held on 3rd January, 1952 at the Indian Science Congress, Calcutta.**

The meeting was attended by twelve members and two visitors. Rev. Father H. Santapau was unanimously voted to take the chair.

In the absence of the Secretary, the minutes of the last Annual General Meeting were read by Dr. B. L. Chona and confirmed. Dr. Chona read the report of the Secretary for the year 1951 and presented the accounts.

While discussing the financial position of the Society, it was found that the heavy cost of the printing was telling upon the resources of the Society. Rev. Father Santapau while appreciating the services of the outgoing President and of the Secretary-Treasurer and the high standard of the Journal in respect of matter and its presentation, suggested that it should be possible to entrust the work to a cheaper press without affecting the quality. The general feeling was that the Editor-in-Chief should contact Father Santapau for this purpose. Dr. P. N. Nandi also said that some presses in Calcutta could take up this work at a low cost.

It was suggested by Prof. R. K. Saxena that the members defaulting for two years should be requested to pay up the arrears. If they fail to pay, their membership should be considered to have terminated and the supply of the Society's publications should be stopped. The feeling of those present was that this should not be enforced for the present because the publication of the Journal has not been regular. For the same reason the question of enhancing the subscription and membership rates was put off.

The resolution moved by Dr. M. K. Patel extending the life of office bearers for three years was not adopted and it was decided to continue the present practice of holding fresh elections every year.

The ballot papers were scrutinised and counted by Dr. K. S. Bhargava and Dr. M. J. Thirumalachar and the following were declared elected office bearers for 1952 :

President	..	..	Dr. B. B. Mundkur.
Vice-President	..	..	Dr. R. P. Asthana.
Councillors	..	..	Dr. A. P. Misra.
			Dr. M. L. Gattani.
			Prof. S. R. Bose.
			Mr. K. M. Thomas.
			Dr. M. K. Patel.
			Dr. P. R. Mehta.

It was felt that the President of the Society should try to attend the Annual General Meetings and deliver the Presidential address.

Members recorded with thanks their appreciation of the services of the outgoing office bearers of the Society. The meeting terminated with votes of thanks to the chair and to Dr. Chona who acted on behalf of the Secretary at the meeting in his absence.



### Book Review

MANUAL OF BACTERIAL PLANT PATHOGENS. 2nd Edition—C. Elliott, Ph.D., Chronica Botanica Company, Waltham, Mass., U.S.A., Macmillan & Co. Calcutta. viii + 186 pages. 6.00.

The book is divided into three parts. In part I are described about 200 plant pathogens belonging to six genera with short description of each. Part II lists about 100 organisms not recognised as valid plant pathogens because of doubtful pathogenicity to plants or those incompletely described. At the end of the volume are given indices of (i) host plants and of (ii) genera and species of pathogens.

Synonyms, morphological and physiological characters, list of hosts, symptoms, geographical distribution and control are given for each organism. References given in the 1930 edition have been amplified and brought upto date by addition of more recent references.

The author who has spent her lifetime almost entirely in investigations on bacterial diseases of plants under the able guidance of that eminent bacteriologist, late E.F. Smith is certainly the right person to compile results on all bacterial plant pathogens. She deserves the gratitude of those working in this field for incorporating all the available references upto 1948 and for adopting Bergey's classification, which according to her is a real progress towards a more uniform and stable nomenclature. Specially to students of this group in India where this phase of plant pathology is in its infancy, this is a welcome volume since it gives all the available information. The reviewer is particularly happy since investigations into phytopathogenic bacteria (30 n.spp. and 10 old ones) have been completed in his laboratory besides establishing a school to investigate further into the bacterial diseases of plants in India which according to Smith was *terra incognita* upto now. The book should, therefore, find a place in all agricultural college libraries and every student of phytopathogenic bacteriology should possess a copy for ready reference.

As in other publications of this Company, the present volume has a nice get up and is carefully printed without any errors.

M.K. Patel.

## FAO Plant Protection Bulletin

*FAO Plant Protection Bulletin, a monthly published by the Food and Agriculture Organization of the United Nations, Rome, Italy. Editor Dr. Lee Ling, Production Branch, Agriculture Division. F.A.O., Rome, Italy. Subscription Rates Rs. 10/- per annum (for Government Departments Rs. 5/- per annum.)*

The Food and Agriculture Organisation of the United Nations have started the publication of the Plant Protection Bulletin which will serve as a medium for the World Reporting Service on the plant pests and diseases. Undoubtedly this publication will be welcomed as it serves a long felt need for the exchange of information on the incidence of plant pests and diseases of economic importance among different countries. The progress of campaign in various countries is also reviewed. There is hardly any need for one to add that international co-operation for control of pests is essential and in this respect the FAO Plant Protection Bulletin will play an important part.

The articles that have appeared in the numbers of the Bulletin issued so far, are of high standard and contain extremely valuable information. The latest Plant Quarantine announcements of different countries published from time to time are also of very great topical interest to Plant Quarantine officials of different countries. The get up of the bulletin is excellent and speaks highly of its editor, Dr. Lee Ling. Much of the success of this Bulletin will depend upon the close cooperation of all member countries of the FAO and in particular the signatories of the International Plant Protection Convention. We are sure all countries will most willingly send the required information to the editor of the Bulletin. In India, the Directorate of Plant Protection, Quarantine and Storage of the Ministry of Food and Agriculture, Government of India is already collating and collecting such information in cooperation with the State Plant Protection Organizations. The summaries will be transmitted to the FAO.

We hope and wish that the FAO Plant Protection Bulletin will grow in its usefulness and be of service to all countries.

H. S. PRUTHI